DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service
Food and Drug Administration

Memorandum

Date AUG 4 2000

From (Acting) Division Director, Division of Standards and Labeling Regulations,
Office of Nutritional Products, Labeling and Dietary Supplements, HFS-820

Subject 75-Day Premarket Notification of New Dietary Ingredients

To Dockets Management Branch, HFA-305

New Dietary Ingredient:	extract of *Agaricus blazei* Merrill
Firm:	Iwade Research Institute of Mycology Co., Inc.
Date Received by FDA:	May 23, 2000
90-Day Date:	August 20, 2000

In accordance with the requirements of section 413(a) of the Federal Food, Drug and Cosmetic Act, the attached 75-day premarket notification for the aforementioned new dietary ingredient should be placed on public display in Docket No. 95S-0316 after August 20, 2000.

Felicia B. Satchell

Felicia B. Satchell

95S-0316

RPT76

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
Washington DC 20204

AUG 4 2000

Kristi O. Smedley, Ph.D.
Consultant
Center for Regulatory Services
5200 Wolf Run Shoals Road
Woodbridge, Virginia 22192

Dear Dr. Smedley:

This is in response to your letter submitted on behalf of Iwade Research Institute of Mycology Company, Inc. of Suehiro-cho, Tsu, Mie, Japan (client) to the Food and Drug Administration (FDA) dated May 22, 2000, making a submission for a new dietary ingredient pursuant to 21 U.S.C. 350b(a)(2) (section 413(a)(2) of the Federal Food, Drug, and Cosmetic Act (the Act)). Your letter notified FDA of your client's intent to market a dietary supplement product containing A new dietary ingredient, namely, an extract of *Agaricus blazei* Murrill. This new dietary ingredient notification contains information that supplements that contained in a previous submission dated May 18, 1999. We concluded in our letter dated July 29, 1999, that the information in the previous submission did not provide a basis to conclude that a dietary supplement containing this new dietary ingredient will reasonably be expected to be safe.

21 U.S.C. 350b(a)(2) requires that a manufacturer or distributor of a dietary supplement that contains a new dietary ingredient submit to FDA, at least 75 days before the dietary ingredient is introduced or delivered for introduction into interstate commerce, information that is the basis on which the manufacturer or distributor has concluded that a dietary supplement containing such new dietary ingredient will reasonably be expected to be safe. FDA reviews this information to determine whether it provides an adequate basis for such a conclusion. Under section 350b(a)(2), there must be a history of use or other evidence of safety establishing that the dietary ingredient, when used under the conditions recommended or suggested in the labeling of the dietary supplement, will reasonably be expected to be safe. If this requirement is not met, the dietary supplement is deemed to be adulterated under 21 U.S.C. 342(f)(1)(b) because there is inadequate information to provide reasonable assurance that the new dietary ingredients do not present a significant or unreasonable risk of illness or injury.

FDA has carefully considered the information in your submission, and the agency has significant concerns about the evidence on which you rely to support your conclusion that the new dietary ingredients stated above will reasonably be expected to be safe. In our letter of July 29, 1999, we stated that there was a lack of quantitative estimates of dietary exposure to *Agaricus blazei* Murrill extract (ABME) that would provide a basis to support the history of use of this substance in Japan to conclude that its use in a dietary supplement is safe. The current submission states that your client "is not requesting that

FDA make a determination of safety based on historical use." Since the history of use will not be used as a basis to conclude that this new dietary ingredient will reasonably be expected to be safe, we have not considered prior human food use of the ingredient in our review of your notification.

Your client's submission contained data from two animal studies and three human studies that your client asserts support a determination that the dietary supplement ABME will reasonably be expected to be safe. All animal studies were performed using adult rodents.

Your client's submission included, in Attachment A, a derived tolerable daily intake (TDI) for "Himematsutake powder" based on the findings of animal studies. Several issues need to be addressed to clarify the basis of your calculations in deriving the TDI. First, no references are noted for the studies cited in Attachment A as the basis for the calculations. Second, it appears the animals used in the studies that were the basis of the TDI (section 4.B.I and 4.B.II) were administered ABME. Thus, a TDI derived from these animal studies represents a TDI for ABME, not Himematsukate powder. In turn, the TDI for ABME cannot be directly compared in a meaningful way to the doses of exposure to Himematsukate powder in human studies as is done in Attachment A. Himematsukate powder is indicated in the submission to contain ABME extract and guar gum. These differences need to be considered in the estimates. This section also indicated that the result of chronic toxicity studies (6-month rat and mouse studies) demonstrated a no adverse effect level (NOAEL) of 3000 mg/kg body weight (bw) and your client derived a TDI from this NOAEL of 30 mg/kg bw/day using an uncertainty factor (UF) of 100 (10 X 10 for intra- and inter-species differences). However, the rat study that was indicated as having a NOAEL of 3000 mg/kg body weight revealed small increases in liver weight expressed as g per 100g body weight in males at 3000 mg ABME/kg bw/day dose level at week 13 and 26, and in females at the 3000 mg/kg bw/day dose level at 13 weeks. The authors of the study suggest that the effect on liver weight is not due to the test material, but the pattern and consistency of this effect suggests that it is ABME-treatment induced. Clarification of the statistical analyses of these changes is warranted. Other changes such as increase in food intake and decrease in cholesterol were seen at 1000 and 3000 mg ABME /kg bw/day in the rat study submitted. It appears that your client concluded that it is reasonable not to consider these changes as adverse effects. If the alteration in liver weight represents an adverse effect, then the lowest adverse effect level (LOAEL) would be 3000 mg/kg bw/day and the NOAEL would instead be 1000 mg/kg bw/day (or possibly 50 mg/kg bw/day based on the mouse study). Then the TDI for ABME based on animal studies would be lower than suggested (e.g., 10 mg/kg bw/day). Finally, in the TDI derivation, your client notes that "the formulated product" administered to healthy humans was 3X-6X the recommended dosage (4500 – 8000 mg/person/day). It is not clear to which study this refers.

Three studies performed in adult humans was also provided in the notification. In the first study (Section 4.B.III), the AMBE used was confirmed as identical to the AMBE

dietary supplement product that Iwade intends to market in the United States (U.S.) (see letter in Section 4.B. III). However, interpretation of the information presented in the letter is difficult. It is not clear if it indicates that the ABME used in the study is identical to ABME used in the Iwade dietary supplement or the ABME used in the study actually represents the Himematsutake powder which contains a diluent and/or is identical to the Iwade dietary supplement product. In addition, the volume (ml) of ABME fluid administered is indicated in the study but the concentration of ABME in this fluid is not noted. Without information on the dose of exposure, it is difficult to draw conclusions about the significance of the paucity of substantial ABME-induced changes indicated for a range of measures in this experiment. Also with respect to this study, the results on these various measures were presented for each individual. However, no summary data were provided nor were statistical analyses performed. Some individual changes or trends were noted. However, the significance of these changes associated with ABME exposure were not clearly delineated or addressed. Considerations of the response of the subset of individuals with pre-existing medical conditions (hypertension, diabetes and high triglycerides, hyper-triglycerides and lipidemia) with respect to the findings from the healthy subjects may also be of concern.

Another human study (Section 4.B.V) involved 10 female patients with cancers of the reproductive system (malignant tumors of the uterus, cervix, ovaries). Some of the subjects underwent a surgical operation (8/10), chemotherapy (1/10) and/or radiotherapy (7/10) prior to the administration of Himematsutake powder (indication that the Himematsutake powder is identical in nature to the Iwade proposed dietary supplement product is not noted). Studies in seriously ill patients that are confounded with different medical conditions, different degrees and types of cancer, and different treatments are of limited utility in evaluating safety of a substance in healthy people. Changes in the immune system along with blood and liver measures were seen with Himematsutake powder intake. The nature and significance of these potential effects being elicited chronically in normal, healthy individuals consuming Himematsutake-based products have not been addressed. Therefore, this human study provides little support for concluding that chronic or long-term consumption of dietary supplements containing ABME will reasonable be expected to be safe in healthy people.

In the third human study (Section 4.B.IV), 20 healthy male and 15 healthy female university student volunteers (19-23 years old , no body weight provided) consumed 30 and 15 g Himematsutake powder per day, respectively, for 6 months. It is reported by the investigator of this study that no significant side effects were observed in this study. In contrast to this statement, examination of Table 3 and 4 in Section 4.B.II suggests some side effects emerged with exposure to Himematsutake powder such as changes in appetite, digestion, general condition, etc. However, exact interpretation of these tables is difficult because many table elements are not clearly labeled or explained. Clarification on the nature of the changes would be useful.

If adequate human data are available, a toxicological–based safety/risk assessment approach should utilize these data to derive a human TDI with estimates from animal work to support it. Some of the human studies presented in this notification could potentially be addressed in this manner. However, deficiencies and uncertainties exist in the information provided in the human studies and in the notification on the Himematsutake powder utilized in the various experiments, i.e., the Iwade dietary supplement (i.e., 1.5g ABME / 3.5 g guar gum), the powder described in Section 4.B.VI (no % ABME to diluent information provided), and how they compare. This information is vital for determining the merits of the arguments made by your client on the safe use of the Himematsutake dietary supplement product. Furthermore, the information you submitted does not address the safety of use of ABME in children or developing animals.

For the reasons discussed above, the information in your submission does not provide an adequate basis to conclude that extract of *Agaricus blazei* Murrill, when used under the conditions recommended or suggested in the labeling of your client's products, will reasonably be expected to be safe in adults or children. Therefore, the products may be adulterated under 21 U.S.C. 342(f)(1)(B) as dietary supplements that contain the new dietary ingredient specified for which there is inadequate information to provide reasonable assurance that such ingredient does not present a significant or unreasonable risk of illness or injury. Introduction of such products into interstate commerce is prohibited under 21 U.S.C. 331(a) and (v).

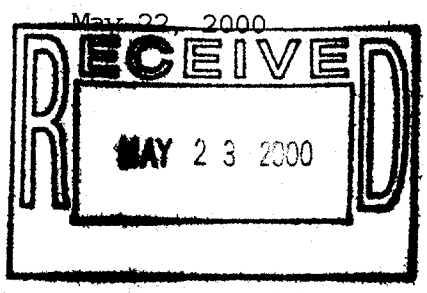
Please contact us if you have any questions concerning this matter.

Sincerely yours,

Felicia B. Satchell

Felicia B. Satchell
(Acting) Division Director
Division of Standards
and Labeling Regulation
Office of Nutritional Products, Labeling
and Dietary Supplement

center for regulatory services
5200 Wolf Run Shoals Road * Woodbridge, VA 22192 * 703 590 7337 * Fax 703 580 8637 * cfrsrv@aol.com



Dr. Robert Moore
Director, Office of Special Nutritionals (HFS-450)
Center for Food Safety and Applied Nutrition
Food and Drug Administration
200 C Street SW
Washington, DC 20204

Dear Dr. Moore:

SUBJECT: Premarket Notification of a New Dietary Ingredient Extract of *Agaricus blazei*-- **SUPPLEMENTAL** Information

On behalf of our client, Iwade Research Institute of Mycology Co., Ltd. (Iwade), notice is hereby given pursuant to the requirements of section 413(a)(2) of the Federal Food, Drug, and Cosmetic Act (21 USC §350b) of the intent of Iwade to introduce into interstate commerce in 75 days herefrom a new dietary ingredient, extract of *Agaricus blazei*. This information is provided in addition to the information submitted on May 18, 1999, and responded to by the agency on July 29, 1999. In accordance with 21 CFR §190.6, enclosed is one original plus two copies of the following information.

We understood that the agency had four concerns regarding the notification submitted on behalf of Iwade: 1) lack of historical quantitative data on consumption; 2) inadequate information about the nature and composition of the extract used in the studies to demonstrate safety; 3) safety information did not support the requested level of supplementation; and 4) the agency requested additional human studies using healthy subjects.

Iwade is not requesting FDA make a determination of safety based on historical use; therefore, we have not addressed that concern. The other concerns are addressed below and in the attached studies.

Iwade has modified the labeling on the product to be used at a level of one package per day (a total of 1.5 grams of Himematsutake extract). We have calculated an NOAEL of 1800 mg; therefore the suggested dose is below the NOAEL (Attachment A).

Information cited under Section 4.B. includes the new information submitted by Iwade to support their determination of safety. You will note that toxicity data referred to by 4.B. I. (Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME" Administered Orally in Rats for 26 weeks) and 4.B.II. (Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME"

Administered Orally in Mice for 26 weeks) were completed by Mie University School of Medicine, as was the complete analysis of the Himematsutake. Also included for your review are two studies completed with healthy volunteers (a 12-week and a 6-month study). Iwade Research has provided a letter of confirmation that the 12-week study was completed with the identical material covered by the notification. The chemical analysis of the product used in the 6 month studies are provided (Attachment 4.B.I.).

1. Manufacture
   Iwade Research Institute of Mycology Co., Ltd.
   1-9, Suehiro-cho, Tsu, Mie
   514-0012, JAPAN

2. New Dietary Ingredient
   Extract of *Agaricus blazei* Murrill (Himematsutake extract)

3. Description Dietary Supplement
   Concentration of the hydrolysis of the culture of *Agaricus blazei*

   > It will be marketed in packages (of Himematsutake) with directions to take orally after dissolving in tepid water.

   > Directions will suggest using one package each day on an empty stomach.

4. Iwade has concluded that the dietary supplement containing Himematsutake extract will reasonably be expected to be safe under the recommended conditions of use based on numerous studies and other information.

   A. Previously Iwade provided the following documents and they are not included again in this filing.

      I. List of Existing Food Additives, Japanese Government (excerpt listing Himematsutake extract and enzymatically hydrolyzed guar gum, English translation and original Japanese)

      II. Summary of Acute and Subacute Toxicological Studies of ABME from Cultured *Agaricus blazei* Murrill (Iwade Strain 101). Hitoshi Ito, M.D. Ph.D., Department of Pharmacology, MIE University School of Medicine, JAPAN (full reports available to FDA).

      III. History of Himematsutake (*Agaricus blazei* Murrill). Iwade Research Institute of Mycology

IV. AGARICUS in North America: Type Studies. Alice E.H. Freeman. 1979. Mycotaxon 8:1.

V. Clinical studies conducted with *Agaricus blazei* indicating no safety problems with the extract:

a. Observation on the Treatment of *Agaricus blazei* for Chronic Hepatitis B. Wang Li Rong et al. Journal of Lanzhou Medical College. Vol. 20. 1994 (English translation and original Japanese)

b. Observation on Treatment Effect of *Agaricus blazei* against Alimentary Tract Tumor. Wang Jing, Mao Xin Min, Cheng Ru Zheng, Wang Jun Zhi, Hitoshi Ito, and Keishiro Shimaru. Gansu Medical Journal. 1994. (English translation and original Japanese)

c. Antitumor Activity and Some Properties of Water-soluble Polysaccharides from "Himematsutake," the Fruiting Body of Agaricus blazei Murrill. Takaishi Mizuno, Toshihiko Hagiwara, et al. Agricultural and Biological Chemistry, 54:2889. 1990.

d. Antitumor Activity and Some Properties of Water-insoluble Hetero-glycans from "Himematsutake," the Fruiting Body of Agaricus blazei Murrill. Takashi Mizuno, Ryuichi Inagaki, et al. Agriculture and Biological Chemistry, 54:2897-2905. 1990.

VI. Manufacturing Scheme **(CONFIDENTIAL)**

VII. Product specifications of Himematsutake Powder and Himematsutake Extract **(CONFIDENTIAL)**

B. In this filing Iwade is providing additional information in support of their determination that the dietary supplement containing Himematsutake extract will reasonably be expected to be safe under the recommended conditions of use.

I. Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME" Administered Orally in Rats for 26 weeks.

II. Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME" Administered Orally in Mice for 26 weeks.

III. Safety of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, ABME, for Humans in Relatively Long Term Oral Administration.

IV Safety Test for Long-term Administration of Himematsutake (Iwade Strain 101) Powder in Healthy Volunteers.

V. Clinical Trail with Himematsutake (Iwade Strain 101) Powder on Patients with Malignant Tumor (Study on Long-Term Administration and Side Effect.

VI. Revised Product specifications of Himematsutake Powder. **(CONFIDENTIAL)**

Should you have any questions or comments on this request, please contact the undersigned.

Sincerely,

Kristi O. Smedley, Ph.D.
Consultant

Enclosures
Listed Above and
on Attachment Page

cc: I. Iwai

506:\043.fda

ATTACHMENTS

A. Tolerable Daily Intake Estaimte – Himematsutake Powder

4.B. I. Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME" Administered Orally in Rats for 26 weeks.

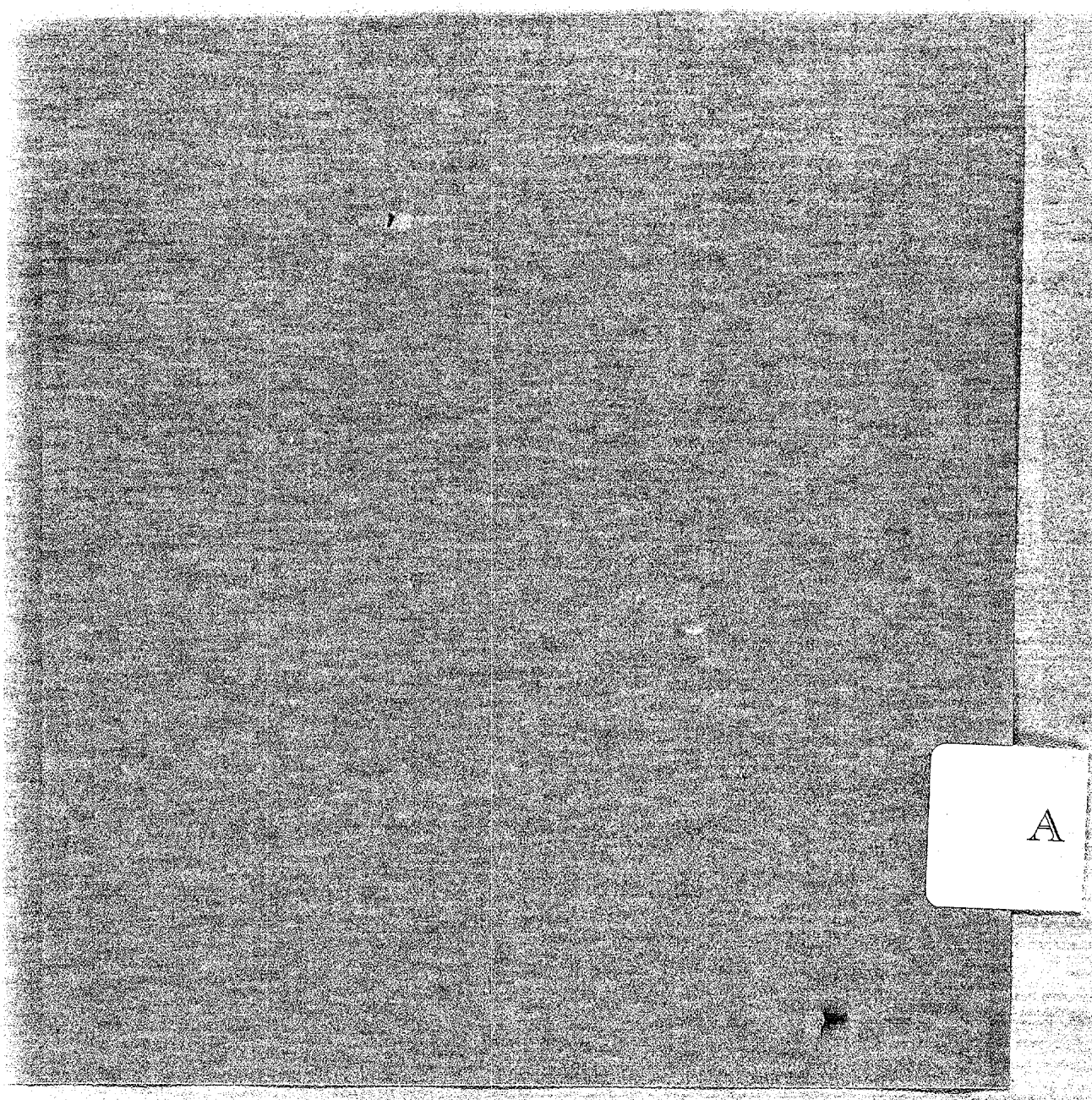
4.B. II. Chronic Study of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, "ABME" Administered Orally in Mice for 26 weeks.

4.B.III. Safety of Cultured Agaricus blazei Murrill (Iwade Strain 101) (Japanese name; Himematsutake) Preparation, ABME, for Humans in Relatively Long Term Oral Administration.

4.B. IV. Safety Test for Long-term Administration of Himematsutake (Iwade Strain 101) Powder in Healthy Volunteers.

4.B. V. Clinical Trial with Himematsutake (Iwade Strain 101) Powder on Patients with Malignant Tumor (Study on Long-Term Administration and Side Effect).

4.B. VI. Revised Product Specifications of Himematsutake Powder. **(CONFIDENTIAL)**


A

Attachı

TOLERABLE DAILY INTAKE ESTIMATE

Himematsutake Powder

The Chronic Toxicity Studies (6-month rat and mouse studies) determined a no adverse effe level (NOAEL) of 3000 mg/kg.

Also, studies of healthy volunteers were administered with the formulated product at levels o to 6x the recommended dosage (4500 mg/person/day - females and 9000 mg/person/day - ma

Consideration of both the chronic toxicity studies in laboratory animals and the lack of a toxic effect in healthy volunteers when administered Himematsutake Powder at 3 or 6x the recommended consumption, it would be appropriate to apply an uncertainty factor of 100 (a factor of 10 for inter-species differences and a factor of 10 for intra-species differences, ie., extrapolation of animal data to human data).

The tolerable daily intake would be 30 mg/kg/day.

With a 60 kg person the tolerable daily intake would be 1800 mg/person/day.

The recommended use is 1500 mg/person/day, thus, the recommended use is under the tolerable daily intake.

506/043a.fda

4.B. I

CHRONIC TOXICITY STUDY OF CULTURED *AGARICUS BLAZEI* MURRILL (IWADE STRAIN 101) [JAPANEASE NAME ; HIMEMATSUTAKE] PREPARATION, "ABME" ADMINISTERED ORALLY IN RATS AND MICE FOR 26 WEEKS.

Hitoshi Ito, M.D., Ph.D. and Keishiro Shimura, M.D.*

Department of Pharmacology Mie University School of Medicine
*Institute of Laboratory Animals, Mie University School of Medicine
2-174, Edobashi, Tsu, Mie 514-0001, Japan

DEPARTMENT OF PHARMACOLOGY
MIE UNIVERSITY SCHOOL OF MEDICINE
EDOBASHI, TSU, MIE 514, JAPAN

Analysis of Experimental Material

Requested by Iwade Research Institute of Mycology, Japan, toxicity studies on ABME with mice and rats were performed. The composition of ABME (cultured *Agaricus blazei* Murrill Extracts) analyzed are as follows;

Material: Himematsutake extract
[ABME : Cultured *Agaricus blazei* Murrill(Iwade strain 101) Extracts]

Description: Himematsutake extract (ABME) is obtained as follows:
Cultured *Agaricus blazei* Murrill(Iwade strain 101) " Himematsutake " washed with distilled water, disintegrated in a mixer, and extracted with boiling water for 5 hours. The suspension was filtered to remove the insoluble material. After concentrating the aqueous extract under reduced pressure, and then spray drying it.

Analytical results

Chemical Specifications

Water Content	*1	g/100g	1.2
Crude ash	*2	g/100g	1.2
Crude protein	*3	g/100g	7.0
Crude fat	*4	g/100g	0.6
Crude fiber	*5	g/100g	0.9
Total sugar	*6	g/100g	19.1

*1 Heat-drying method, 105℃ 3hr
*2 Ashnized method, 550℃ (Carbonizing)
*3 Lowry method
*4 Ether extracting method
*5 Henneberg-Stohmann modified method
*6 Phenol-Sulfuric acid method

DEPARTMENT OF PHARMACOLOGY
MIE UNIVERSITY SCHOOL OF MEDICINE
EDOBASHI, TSU, MIE 514, JAPAN

Amino acid Profile

Aspartic acid	mg/100g	236
Threonine	mg/100g	136
Serine	mg/100g	129
Glutamic acid	mg/100g	230
Glycine	mg/100g	194
Alanine	mg/100g	208
Valine	mg/100g	135
Methionine	mg/100g	36
Leucine	mg/100g	216
Tyrosine	mg/100g	60
Phenylalanine	mg/100g	107
Histidine	mg/100g	57
Lysine	mg/100g	143
Arginine	mg/100g	291
Isoleucine	mg/100g	59
Proline	mg/100g	62

Amino acid analyser

Carbohydrates Profile

Glucose	g/100g	4.9
Galactose	g/100g	2.2
Mannose	g/100g	10.0
Xylose	g/100g	0.2
Arabinose	g/100g	0.06
Ribose	g/100g	1.5
Fucose	g/100g	Trace
Unknown	g/100g	0.27

GLC: gas liquid chromatography

Polysaccharide Profile

β-Glucan	p/100g	7.5
α-Glucan	p/100g	2.2
β-Glucomannan	p/100g	8.4
β-Galactogulucan	p/100g	2.2
Ribonucleotide	p/100g	2.2
Protein bound・β-Glucan	p/100g	8.6
Xyloglucan	p/100g	1.1

^{13}C-NMRanalysis

Two-dimensional COSY analysis

Chronic Toxicity Study of Cultured *Agaricus blazei* Murrill (Iwade Strain 101) (Japanease name ; Himematsutake) Preparation, "ABME" Administered Orally in Rats for 26 Weeks.

Hitoshi Ito, M.D., Ph.D. and Keishiro Shimura, M.D.*

Department of Pharmacology Mie University School of Medicine
*Institute of Laboratory Animals, Mie University School of Medicine
2-174, Edobashi, Tsu, Mie 514-0001, Japan

Introduction

A chronic toxicity study of the edible mushroom, *Agaricus blazei* Murrill (Japanese name; Himematsutake) preparation, "ABME" - *Agaricus blazei* Murrill Extract, Japanese name: Himematsutake, was carried out with Sprague-Dawley / SLC (SD) rats. The ABME was administered orally for 26 weeks in doses of 0, 1000 and 3000 mg/kg/day.

Based on the series of animal experiments studied for the antitumor effect of ABME, the usual dose for human is estimated 25mg/kg. The chronic toxicity study on rats in this report includes 1000mg/kg - 40 times and 3000mg/kg - 120 times more dose compared to the usual dose for human.

With the limitation of the capacity of a rat's stomach and the physical condition of ABME in mind, over 3000mg/kg dose to a rat would be impossible.

ABME was provided by Iwade Research Institute of Mycology, Japan.

Chronic toxicity studies

Animals employed were SD strain rats (Japan SLC, Inc.) The animals were housed and fed in an animal room of the temperature of 23±2℃ and the humidity of 55±5%. Each animal was given solid diet (CLEA Japan CE-2) and water ad libitum.

One group of animals was used of 10 males and 10 females. Doses of administration were determined by the results of subacute toxicity studies, and two grades were adopted; 1000 and 3000 mg/kg/day (The maximum dose are able to the oral administration).

Test materials are easily soluble in water but high concentration used the state of suspensions. Their water solutions were, therefore, prepared as to be at a level of 1000 and 3000 mg/kg of rats body weight. They were compulsorily administered with a gastric catheter of teflon orally. After the test materials were administered, general symptoms of animals were observed for every day.

Results

(1) Behavior

In rat administered orally with 1000 and 3000 mg/kg for 26 weeks, any abnormal findings that seemed to be caused by the administration of the test material were not observed.

(2) Body Weight Changes (Table 1 and Table 2)

The animals were weighed weekly. No inhibition of body weight gain was found during the periods of the experiments among the test animals, both male and female.

(3) Amount of Diet Ingested (Table 3 and Table 4)

The amount of diet ingested weekly per head in every group is as shown Table 3 and Table 4. Food consumption was slightly increased in the early period (at 2nd and 3rd week) and intermediate period (at 8th and 9th week) of administration in male rats. No significant change was found in female rats, so it was not considered that the testing material caused the diet efficacy.

(4) Findings in Hematological Examinations (Table 5, 6, 7 and Table 8)

Hematological examination was performed on 5 cases of each group. No variation of significance was found in red blood cell count, hematocrit value, hemoglobin content, platelet value and white blood cell count. Differential leukocyte was found by fixing blood smear and staining by May-Grünwald Giemsa method. In differential leukocyte count, no abnormal findings were found due to the administration of the test material.

(5) Biochemical Examination of Blood (Table 9, 10, 11 and Table 12)

Biochemical examinations of blood were performed on 5 cases each of the groups, and results obtained are shown in Table (Table 9 - 12). Total cholesterol values in the male animals of the 3000 mg/kg group at 13th and 26th week and in the female animals of the 1000 mg/kg and 3000 mg/kg groups at 26th week were found with significant decrease. With regard to glucose, urea, total protein, albumin, alkaline phosphatase, GOT, GPT, Na and K content, however, no change was observed.

(6) Findings in Urine (Table 13 and Table 14)

Urine protein was assayed in the concentration of trace to 100mg/dl in most of the groups, regardless of the administered or the control. Inspecting urine volume, pH, specific gravity, urobilinogen, bilirubin, ketone body and glucose, no abnormal data was found in all groups.

(7) Findings at Autopsy Organ Weight (Table 15, 16, 17 and Table 18)

A slight increasing tendency was observed in the liver of male groups with administered 3000mg/kg/day at 13 weeks and 26 weeks, and in the female group with administered 3000 mg/kg/day at 13 weeks. However, no remarkable change was found between the control group and the treated groups in either absolute organ weight or comparative organ weight. The changes found were not considered to be caused by the test material.

(8) Histopathological Findings (PHOTO 1—PHOTO 13)

After autopsy and gross observation of changes, the organs were fixed with 10% formalin, embedded in paraffin and cut in slices ca. 6 μ thick, then stained with hematoxylin and eosine. Bone marrows were decalcified by dipping them in 5% nitric acid (10% formalin) for 48 hours.

Microscopic examinations were performed on 5 samples each of the groups at the end of 13 weeks and 26weeks after the administration. Histopathological examination was conducted by Sensake Naruse, M.D., at Department of Pathology, Mie University School of Medicine, Tsu, Mie, 514-0001, Japan.

Lungs : Tuberous infiltrations of cells composed mainly of lymphocytes were seen around the blood vessels in almost all cases including those of the control group.

Liver : Almost no difference between the control and the administered groups; a slight degeneration of liver and enlargement at sinus were observed in 1 case of the control group.

Kidneys : Congestions of glomeruli and slight degenerations of the epithelium of tubules were observed both in the control and the administered groups.

Spleen : A slight hemosiderosis was observed in almost all cases including those of the control group.

* No remarkable changes were observed in brain, heart, testes, ovaries, thymus, pituitary, thyroids, adrenals, pancreas, digestive tracts and bone marrow.

Summary

A chronic toxicity of edible mushroom, *Agaricus blazei* Murrill (Japanese name: Himematsutake) preparation, "ABME" was studied with SD rats.

ABME was administered orally for 26 weeks in dose of 0 (control), 1000 and 3000 mg/kg/day. During the period of oral administration for 26 weeks, no general symptoms to be marked were observed in SD rats, and there was no death throughout the whole period.

With regard to the amount of diet ingested, no significant change was found in all the administered group.

No inhibition of body weight gain was found during the periods of the experiments

among the test animals, both male and female.

In hematological findings, any significant variation in red blood cell count, hematocrit value, hemoglobin content, platelet value and white blood cell count was not found. In differential count, too, no abnormal findings due to the administration of the test material was found.

In biochemical examination of blood, no change was found in glucose, urea, total protein, albumin, alkaline phosphatase, GOT, GPT, Na and K content. However, the significant decrease in total cholesterol values was observed in the male and female of 3000 mg/kg administered groups after 26 weeks.

No abnormality was found in the urine volume, pH, specific gravity, urobilinogen, bilirubin, ketone body, protein and glucose in the control and the administered groups.

In assaying organ weight, a slight tendency of increase of liver was observed in the male and female groups of 3000 mg/kg/day administered group. However, the effect was found to be very slight. With regard to the other organ weight, no change was found in either the administered group of male or female rats compared with the control group.

In the histopathogical examinations, any abnormal figures specific to the administered group compared with the control group was not observed in rats. Furthermore, any toxicity to be caused by ABME could not be found.

Therefore, the safety dose for rats was estimated to be over 3000 mg/kg/day, but the sure intoxication dose could not be determined.

End of report

Table 1 , Body weight changes in male rats given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Male (g)			
	Number of rats	Control	1000	3000
0	15	225	217	210
1	15	268	270	265
2	15	331	328	319
3	15	357	366	358
4	15	401	394	396
5	15	419	415	417
6	15	433	450	448
7	15	451	461	453
8	15	476	472	469
9	15	501	516	507
10	15	519	523	520
11	15	525	530	528
12	15	537	541	536
13	15	542	549	540
14	10	546	552	547
15	10	554	561	557
16	10	561	567	562
17	10	567	569	567
18	10	574	578	575
19	10	587	590	583
20	10	600	607	603
21	10	619	617	615
22	10	623	621	619
23	10	629	627	620
24	10	630	632	626
25	10	632	636	630
26	10	636	642	638

Table 2 Body weight changes in female rats given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Female (g)			
	Number of rats	Control	1000	3000
0	15	164	167	170
1	15	187	189	193
2	15	198	199	204
3	15	213	226	227
4	15	229	238	238
5	15	241	250	249
6	15	263	261	258
7	15	266	267	264
8	15	275	279	278
9	15	281	285	284
10	15	289	290	287
11	15	295	294	293
12	15	298	297	299
13	15	302	304	301
14	10	303	306	304
15	10	305	307	305
16	10	307	312	308
17	10	309	314	311
18	10	315	316	312
19	10	320	319	318
20	10	321	323	320
21	10	323	325	321
22	10	323	326	324
23	10	326	328	324
24	10	326	329	326
25	10	329	332	330
26	10	331	336	334

Table 3 Food consumption of male rats given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Male (g)			
	Number of rats	Control	1000	3000
1	15	26.6±0.5	28.1±0.6	27.8±0.6
2	15	27.9±0.4	29.9±0.5*	29.5±0.6*
3	15	28.4±0.6	31.0±0.8*	30.8±0.7*
4	15	29.9±0.5	31.7±0.7	31.2±0.8
5	15	30.6±0.5	31.4±0.5	31.4±0.9
6	15	30.4±0.5	30.4±0.6	31.3±1.1
7	15	29.5±0.7	30.1±0.7	31.3±1.0
8	15	30.6±1.0	33.2±0.8*	33.0±1.0*
9	15	30.3±0.5	32.0±0.5*	33.1±0.7*
10	15	30.2±0.5	32.1±0.9	31.9±0.8
11	15	30.7±0.4	31.7±0.5	31.5±0.6
12	15	30.5±0.5	33.0±0.5*	31.8±0.9
13	15	30.7±0.5	31.8±0.9	31.1±0.8
14	10	30.8±0.6	31.5±0.8	31.8±0.9
15	10	31.7±1.0	31.0±0.7	31.1±1.0
16	10	33.3±1.1	31.7±0.7	31.9±0.9
17	10	32.1±1.0	31.8±0.6	31.7±0.8
18	10	32.0±1.1	31.5±0.6	31.6±0.9
19	10	31.1±1.3	31.5±0.7	31.7±0.8
20	10	31.5±1.0	31.7±0.7	30.9±1.0
21	10	32.0±1.1	31.1±0.8	31.7±1.1
22	10	31.6±1.0	31.9±0.7	30.6±1.0
23	10	30.8±1.1	30.5±0.6	30.4±0.8
24	10	31.3±0.9	30.9±0.6	30.0±0.9
25	10	31.0±0.9	31.5±0.7	30.8±0.8
26	10	31.0±0.8	32.9±1.3	31.6±0.9

Values represent mean ± standard error (g/day/rat)
* Significantly different from control at $p < 0.05$

Table 4 Food consumption of female rats given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Female (g)			
	Number of rats	Control	1000	3000
1	15	20.5±0.5	19.2±0.5	19.6±0.5
2	15	19.6±0.4	20.1±0.4	20.8±0.5
3	15	19.5±0.5	20.2±0.6	20.9±0.4
4	15	21.2±0.6	20.1±0.7	21.2±0.5
5	15	21.0±0.4	20.2±0.5	21.2±0.6
6	15	21.2±0.5	20.2±0.5	21.0±0.5
7	15	21.0±0.7	20.3±0.4	21.5±0.6
8	15	22.8±0.6	21.7±0.7	22.5±0.5
9	15	22.3±0.4	21.2±0.5	22.6±0.5
10	15	22.5±0.4	21.4±0.8	22.1±0.4
11	15	22.0±0.5	20.7±0.5	19.9±0.9
12	15	22.6±0.5	21.5±0.8	22.3±0.5
13	15	21.8±0.6	21.6±0.3	21.5±0.5
14	10	22.0±0.6	21.2±0.5	21.8±0.4
15	10	22.4±0.6	22.2±0.5	22.4±0.5
16	10	23.8±1.0	22.9±0.9	23.6±0.8
17	10	20.6±0.7	21.2±0.6	21.3±0.6
18	10	20.9±0.6	21.8±0.7	22.0±1.1
19	10	20.2±0.7	21.9±0.9	21.7±0.7
20	10	20.6±0.6	21.0±0.8	21.2±0.6
21	10	20.9±0.5	21.6±0.6	21.4±0.5
22	10	20.5±0.6	20.6±0.7	20.7±0.5
23	10	20.9±0.6	21.2±0.8	20.7±0.7
24	10	21.4±0.8	21.8±0.7	21.1±0.7
25	10	20.5±0.5	21.1±0.7	20.9±0.5
26	10	20.9±0.9	21.0±0.7	20.4±0.7

Values represent mean ± standard error (g/day/rat)
* Significantly different from control at $p < 0.05$

Table 5 Hematological findings in male rats given ABME orally for 13 weeks

Male											
Dose level (mg/kg/day)	Number of rats	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%) [a]				
							L	M	N	E	B
Control	5	857±40.3	44±0.9	15.2±0.3	107±9.3	91±15.2	80.6	4.1	14.7	0.6	0
							(71-89)	(2-5)	(9-23)	(0-1)	
1000	5	861±37.2	45±1.3	14.9±0.4	111±8.4	119±25.3	81.0	3.4	14.9	0.7	0
							(70-89)	(2-6)	(8-26)	(0-2)	
3000	5	860±42.1	46±1.7	15.1±0.2	109±8.1	107±21.1	79.4	3.7	15.9	1.0	0
							(72-84)	(1-5)	(9-30)	(0-2)	

Values represent mean ± standard error [a] Ranges are given parentheses.
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 6 Hematological findings in female rats given ABME orally for 13 weeks

Female											
Dose level (mg/kg/day)	Number of rats	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%) [a]				
							L	M	N	E	B
Control	5	752±30.5	39±1.2	14.2±0.4	94±5.7	73±11.4	81.4	2.7	14.5	1.4	0
							(73-88)	(1-5)	(8-24)	(1-3)	
1000	5	750±29.3	41±1.4	14.8±0.6	97±8.1	85±10.0	82.3	2.6	14.1	1.0	0
							(70-87)	(1-4)	(9-22)	(1-2)	
3000	5	769±26.0	40±1.2	14.5±0.6	92±9.8	91±9.4	82.0	2.8	14.0	1.2	0
							(74-89)	(1-5)	(8-21)	(0-3)	

Values represent mean ± standard error [a] Ranges are given parentheses.
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 7 Hematological findings in male rats given ABME orally for 26 weeks

Male											
Dose level (mg/kg/day)	Number of rats	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%) [a]				
							L	M	N	E	B
Control	10	865±43.1	43±1.0	15.0±0.3	106±9.6	89±11.2	77.9	2.8	17.0	2.3	0
							(62-89)	(0-4)	(6-33)	(1-3)	
1000	10	868±39.7	44±0.9	15.6±0.2	107±8.5	97±13.1	78.8	2.8	15.9	2.5	0
							(64-87)	(1-5)	(7-32)	(1-4)	
3000	10	870±44.2	43±1.2	15.5±0.4	104±9.0	101±15.8	73.9	2.6	21.0	2.5	0
							(60-84)	(0-5)	(8-37)	(1-3)	

Values represent mean ± standard error [a] Ranges are given parentheses.
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 8 Hematological findings in female rats given ABME orally for 26 weeks

Female											
Dose level (mg/kg/day)	Number of rats	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%) [a]				
							L	M	N	E	B
Control	10	769±40.1	41±0.7	15.2±0.3	96±9.9	75±11.0	71.8	1.9	24.9	1.4	0
							(60-83)	(0-4)	(14-39)	(0-2)	
1000	10	771±39.3	44±1.3	14.9±0.2	101±9.1	79±9.5	70.8	2.0	25.2	2.0	0
							(59-81)	(1-4)	(13-41)	(0-3)	
3000	10	760±36.0	43±1.5	15.3±0.4	103±8.7	83±12.1	72.3	2	23.8	1.9	0
							(61-85)	(0-5)	(11-35)	(0-4)	

Values represent mean ± standard error [a] Ranges are given parentheses.
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 9 Biochemical findings in male rats given ABME orally for 13 weeks

Male											
Dose level (mg/kg/day)	Number of rats	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)	Na (mEq/l)	K (mEq/l)
Control	5	192±36	18±2	7.6±0.2	2.6±0.2	262±46.3	61±6.4	30±4.9	80±8.9	141±2	-
1000	5	184±21	19±2	7.9±0.4	2.7±0.1	259±43.9	60±6.0	29±3.7	72±7.3	143±1	-
3000	5	180±22	20±3	8.0±0.4	2.9±0.2	244±52.0	62±8.6	31±5.2	62±5.7*	144±2	-

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method
Na and K : Flame reaction

Table 10 Biochemical findings in female rats given ABME orally for 13 weeks

Female											
Dose level (mg/kg/day)	Number of rats	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)	Na (mEq/l)	K (mEq/l)
Control	5	142±19	20±1	7.9±0.2	3.0±0.5	181±32.6	59±7.8	27±5.5	64±7.2	-	5.4±0.4
1000	5	128±17	19±1	8.1±0.4	3.2±0.3	201±39.3	63±9.3	26±6.1	58±5.8	-	5.9±0.7
3000	5	123±10	18±1	7.8±0.3	2.9±0.2	190±40.2	67±6.5	24±6.8	59±3.2	-	5.2±0.6

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method
Na and K : Flame reaction

Table 11 Biochemical findings in male rats given ABME orally for 26 weeks

Male											
Dose level (mg/kg/day)	Number of rats	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)	Na (mEq/l)	K (mEq/l)
Control	10	170±12	19±3	7.5±0.2	2.7±0.1	293±46.6	69±5.6	46±3.1	92±8.0	145±2	5.1±0.5
1000	10	165±27	18±2	8.0±0.3	2.5±0.2	263±40.3	72±7.3	45±3.0	84±8.1	143±2	4.9±0.4
3000	10	159±16	19±1	7.7±0.2	2.7±0.2	287±24.9	66±9.1	46±6.2	76±5.3*	142±1	4.7±0.5

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method
Na and K : Flame reaction

Table 12 Biochemical findings in female rats given ABME orally for 26 weeks

Female											
Dose level (mg/kg/day)	Number of rats	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)	Na (mEq/l)	K (mEq/l)
Control	10	132±21	20±3	8.2±0.3	3.1±0.4	169±38.5	86±10.1	49±6.2	82±9.1	143±2	4.9±0.5
1000	10	125±13	19±1	8.7±0.3	3.6±0.3	227±29.6	92±9.8	47±7.0	64±5.3*	142±1	5.0±0.4
3000	10	133±9	19±0	7.9±0.2	3.6±0.2	230±32.7	84±5.6	43±7.4	63±6.0*	143±0	4.8±0.3

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method
Na and K : Flame reaction

Table 13 Urinalysis of male rats given ABME orally for 26 weeks

Male											
Dosing period (week)	Dose level (mg/kg/day)	Number of rats	Appearance	Volume (ml)	pH	Specific gravity	Urobilinogen (Ehrlich unit/dl)	Bilirubin	Ketone body	Protein	Glucose
13	Control	10	Normal	12.9±1.4	7.2 (6.8-7.8)[a)]	1.048 (1.004-1.065)	0.1-1	−	−	±~+	−
	1000	10	Normal	11.6±1.0	7.0 (6.7-7.5)	1.063 (1.051-1.074)	0.1-1	−	−	±~+	−
	3000	10	Normal	13.2±1.2	7.4 (6.6-7.7)	1.044 (1.029-1.053)	0.1-1	−	−	±~+	−
26	Control	10	Normal	13.0±1.0	6.7 (6.3-7.0)	1.056 (1.053-1.073)	0.1-1	−	−	±~+	−
	1000	10	Normal	14.1±1.9	6.6 (6.2-7.1)	1.050 (1.039-1.076)	0.1-1	−	−	±~+	−
	3000	10	Normal	12.9±0.9	6.7 (6.1-7.2)	1.051 (1.039-1.060)	0.1-1	−	−	±~+	−

[a)] Ranges are given parentheses.
pH : pH meter,
Specific grarity : Weight determination,
Urobilinogen, Bilirubin, Ketone body,Protein and Glucose : Uro-Labstix (Ames reagent strips for urinalysis)

Table 14 Urinalysis of female rats given ABME orally for 26 weeks

Female											
Dosing period (week)	Dose level (mg/kg/day)	Number of rats	Appearance	Volume (ml)	pH	Specific gravity	Urobilinogen (Ehrlich unit/dl)	Bilirubin	Ketone body	Protein	Glucose
13	Control	10	Normal	10.7±1.5	7.1 (6.9-8.2)[a]	1.044 (1.023-1.063)	0.1-1	−	−	±～+	−
	1000	10	Normal	10.4±1.3	7.0 (6.5-8.7)	1.047 (1.032-1.060)	0.1	−	−	±～+	−
	3000	10	Normal	9.8±1.0	7.4 (6.9-9.0)	1.048 (1.037-1.065)	0.1-1	−	−	±～+	−
26	Control	10	Normal	12.3±2.0	7.0 (6.3-7.1)	1.048 (1.035-1.067)	0.1	−	−	−～+	−
	1000	10	Normal	13.0±1.7	7.3 (6.9-7.7)	1.050 (1.040-1.071)	0.1	−	−	−～+	−
	3000	10	Normal	13.4±1.9	7.1 (6.2-7.6)	1.046 (1.029-1.063)	0.1	−	−	−～+	−

[a] Ranges are given parentheses.
pH : pH meter,
Specific grarity : Weight determination,
Urobilinogen, Bilirubin, Ketone body,Protein and Glucose : Uro-Labstix (Ames reagent strips for urinalysis)

Table 15 Organ weights in male rats given ABME orally for 13 weeks

Male													
Dose level (mg/kg/day)	Number of rats	Final body wt. (g)	Brain (g)	Heart (g)	Lung (g)	Liver (g)	Kidneys (g)	Spleen (g)	Testes (g)	Thymus (g)	Pituitary (mg)	Thyroids (mg)	Adrenals (mg)
Control	5	542±30	1.99±0.02	1.43±0.07	1.83±0.05	14.80±1.21	3.59±0.20	0.78±0.02	3.52±0.10	0.35±0.03	15±1	27±2	62±2
			(0.37±0.01)	(0.26±0.02)	(0.34±0.01)	(2.73±0.06)	(0.66±0.01)	(0.14±0.01)	(0.65±0.04)	(0.065±0.004)	(2.8±0.3)	(4.9±0.7)	(11±1)
1000	5	549±27	1.94±0.04	1.41±0.09	1.85±0.06	15.10±1.07	3.73±0.15	0.80±0.04	3.26±0.19	0.45±0.08	16±2	28±1	64±3
			(0.35±0.03)	(0.26±0.02)	(0.34±0.02)	(2.75±0.07)	(0.68±0.03)	(0.15±0.01)	(0.59±0.06)	(0.082±0.012)	(2.9±0.4)	(5.1±0.3)	(12±1)
3000	5	540±35	2.12±0.04	1.43±0.08	1.86±0.12	15.91±0.79	3.62±0.09	0.82±0.05	3.50±0.09	0.41±0.06	15±2	28±1	61±3
			(0.39±0.02)	(0.26±0.01)	(0.34±0.02)	(2.95±0.06)	(0.67±0.02)	(0.15±0.01)	(0.65±0.04)	(0.076±0.011)	(2.8±0.5)	(5.2±0.2)	(11±1)

Values represent mean ± standard error.
Values in parentheses represent organ weights in grams or milligrames per 100g body weight.

Table 16 Organ weights in female rats given ABME orally for 13 weeks

Female													
Dose level (mg/kg/day)	Number of rats	Final body wt. (g)	Brain (g)	Heart (g)	Lung (g)	Liver (g)	Kidneys (g)	Spleen (g)	Ovaries (mg)	Thymus (g)	Pituitary (mg)	Thyroids (mg)	Adrenals (mg)
Control	5	302±18	1.91±0.03	0.87±0.03	1.37±0.06	8.25±0.40	2.04±0.06	0.52±0.03	72±4	0.31±0.03	16±1	22±2	72±2
			(0.63±0.03)	(0.29±0.01)	(0.45±0.02)	(2.73±0.08)	(0.68±0.02)	(0.17±0.01)	(24±2)	(0.102±0.009)	(5.3±0.3)	(7.3±0.6)	(24±1)
1000	5	304±19	1.88±0.03	0.85±0.04	1.38±0.09	8.40±0.51	1.97±0.06	0.50±0.04	73±4	0.32±0.04	16±2	23±1	69±2
			(0.62±0.04)	(0.28±0.01)	(0.45±0.03)	(2.76±0.07)	(0.65±0.02)	(0.16±0.01)	(24±2)	(0.105±0.015)	(5.3±0.3)	(7.6±0.4)	(23±1)
3000	5	301±16	1.89±0.04	0.90±0.05	1.30±0.07	8.67±0.43	2.01±0.09	0.53±0.04	70±6	0.33±0.02	18±3	23±2	71±3
			(0.63±0.02)	(0.30±0.02)	(0.43±0.04)	(2.88±0.07)	(0.67±0.03)	(0.18±0.01)	(23±3)	(0.110±0.010)	(6.0±0.4)	(7.6±0.4)	(24±1)

Values represent mean ± standard error.
Values in parentheses represent organ weights in grams or milligrames per 100g body weight.

Table 17 Organ weights in male rats given ABME orally for 26 weeks

Male													
Dose level (mg/kg/day)	Number of rats	Final body wt. (g)	Brain (g)	Heart (g)	Lung (g)	Liver (g)	Kidneys (g)	Spleen (g)	Testes (g)	Thymus (g)	Pituitary (mg)	Thyroids (mg)	Adrenals (mg)
Control	10	636±24	2.09±0.04	1.52±0.06	2.16±0.05	16.85±0.90	3.69±0.11	0.93±0.05	3.55±0.26	0.27±0.03	17±1	29±1	60±2
			(0.33±0.01)	(0.24±0.01)	(0.34±0.01)	(2.65±0.07)	(0.58±0.01)	(0.15±0.01)	(0.56±0.03)	(0.043±0.004)	(2.6±0.2)	(4.6±0.2)	(9±0)
1000	10	642±20	2.05±0.03	1.60±0.07	2.25±0.10	17.79±1.11	3.78±0.15	1.05±0.04	3.34±0.15	0.25±0.03	16±0	28±1	61±2
			(0.32±0.01)	(0.25±0.02)	(0.35±0.02)	(2.77±0.07)	(0.59±0.03)	(0.16±0.01)	(0.52±0.03)	(0.039±0.002)	(2.5±0.1)	(4.4±0.2)	(10±1)
3000	10	638±19	2.16±0.04	1.65±0.05	2.25±0.09	18.29±0.81	3.85±0.16	1.00±0.05	3.62±0.21	0.28±0.03	17±1	30±1	60±3
			(0.34±0.01)	(0.26±0.02)	(0.35±0.02)	(2.87±0.08)	(0.60±0.02)	(0.16±0.02)	(0.57±0.03)	(0.044±0.003)	(2.7±0.1)	(4.6±0.2)	(9±0)

Values represent mean ± standard error.
Values in parentheses represent organ weights in grams or milligrames per 100g body weight.

Table 18 Organ weights in female rats given ABME orally for 26 weeks

Female													
Dose level (mg/kg/day)	Number of rats	Final body wt. (g)	Brain (g)	Heart (g)	Lung (g)	Liver (g)	Kidneys (g)	Spleen (g)	Ovaries (mg)	Thymus (g)	Pituitary (mg)	Thyroids (mg)	Adrenals (mg)
Control	10	331±12	1.88±0.03	0.97±0.03	1.53±0.04	8.49±0.30	2.10±0.07	0.57±0.03	57±6	0.17±0.01	24±1	25±1	79±4
			(0.57±0.01)	(0.29±0.01)	(0.46±0.01)	(2.56±0.03)	(0.63±0.02)	(0.17±0.01)	(17±3)	(0.053±0.003)	(7.3±0.5)	(7.6±0.5)	(23±1)
1000	10	336±16	1.86±0.03	1.04±0.05	1.58±0.08	8.91±0.39	2.14±0.06	0.61±0.02	58±5	0.18±0.02	25±2	25±1	82±5
			(0.55±0.02)	(0.31±0.01)	(0.47±0.01)	(2.65±0.07)	(0.64±0.02)	(0.18±0.01)	(17±1)	(0.054±0.004)	(7.4±0.3)	(7.4±0.6)	(24±2)
3000	10	334±13	1.87±0.06	0.95±0.03	1.51±0.07	8.35±0.50	1.99±0.05	0.60±0.03	56±6	0.17±0.02	23±1	25±1	80±3
			(0.56±0.03)	(0.28±0.01)	(0.45±0.02)	(2.50±0.06)	(0.60±0.02)	(0.18±0.01)	(17±1)	(0.051±0.008)	(6.9±0.4)	(7.5±0.3)	(24±1)

Values represent mean ± standard error.
Values in parentheses represent organ weights in grams or milligrames per 100g body weight.

PHOTO 1 **Lungs**: 3000mg/kg/day×26 weeks, male, ×100,
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

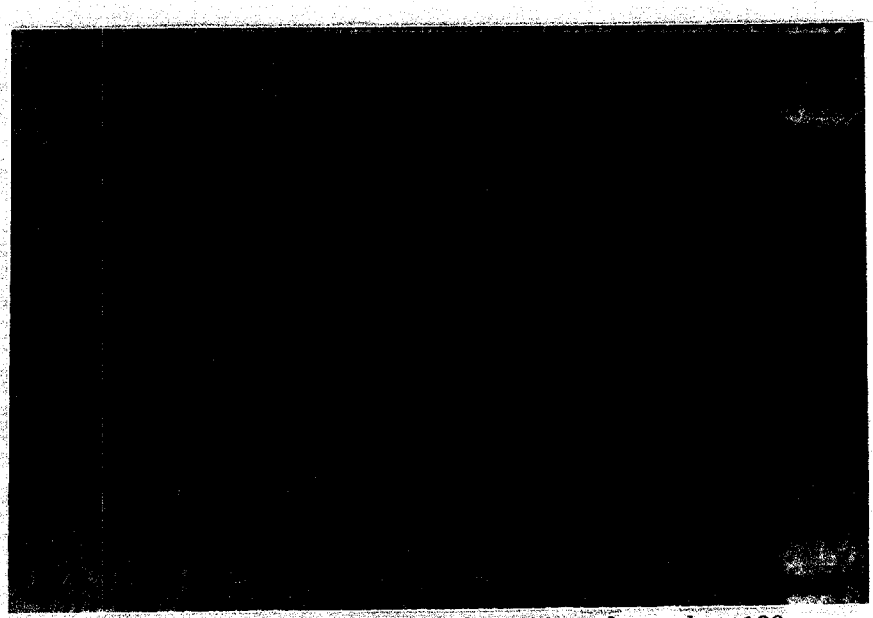


PHOTO 2 **Spleen**: 3000mg/kg/day×26 weeks, male, ×100
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

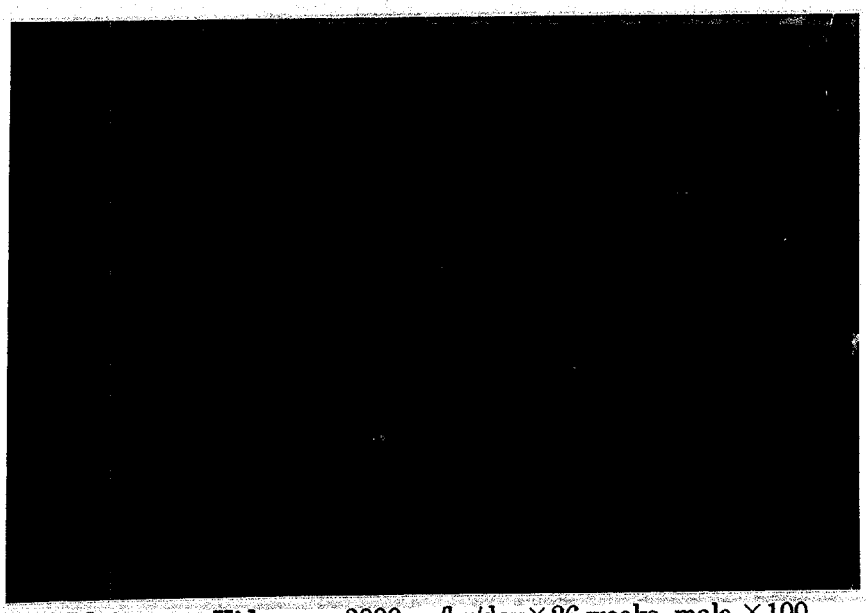


PHOTO 3 **Kidneys**: 3000mg/kg/day×26 weeks, male, ×100,
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

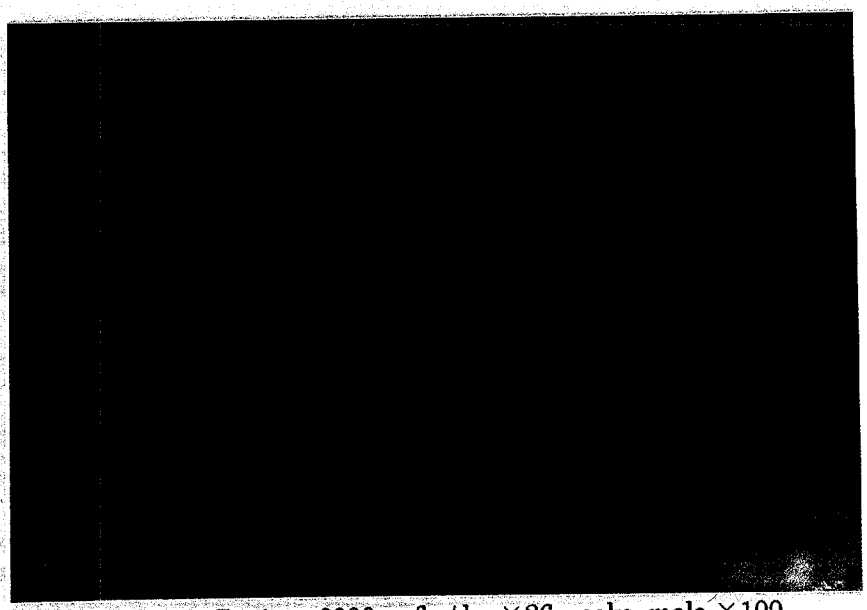


PHOTO 4 **Brain**: 3000mg/kg/day×26 weeks, male, ×100.
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

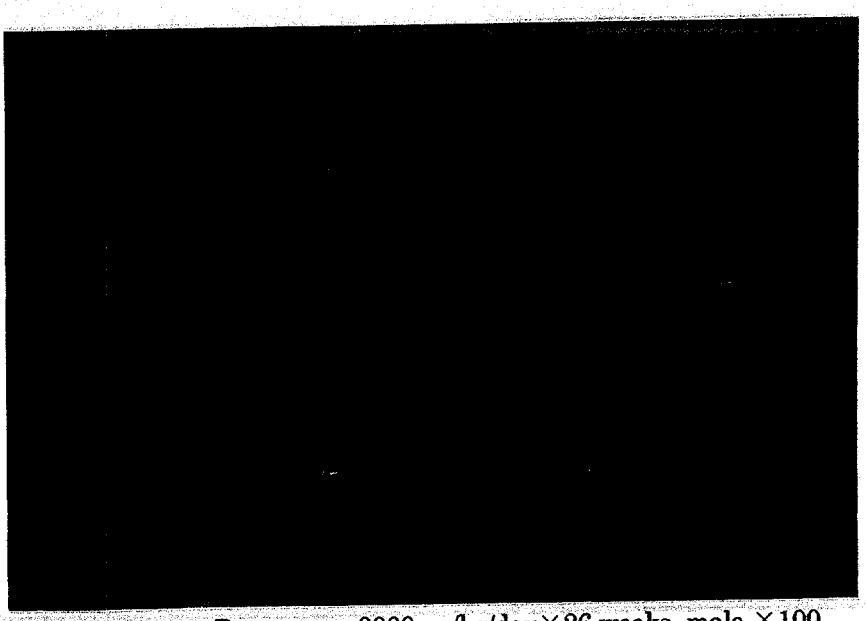


PHOTO 5 **Pancreas:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

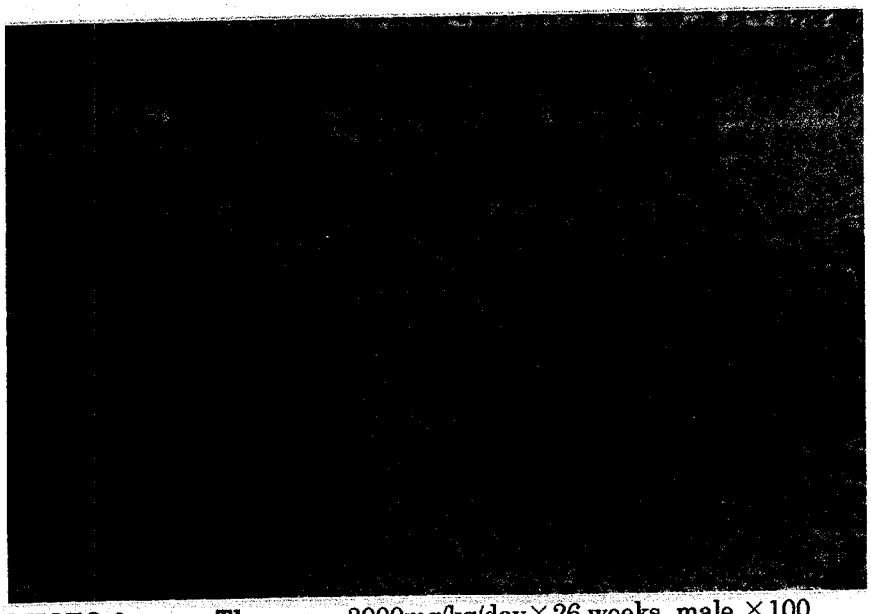


PHOTO 6 **Thymus:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

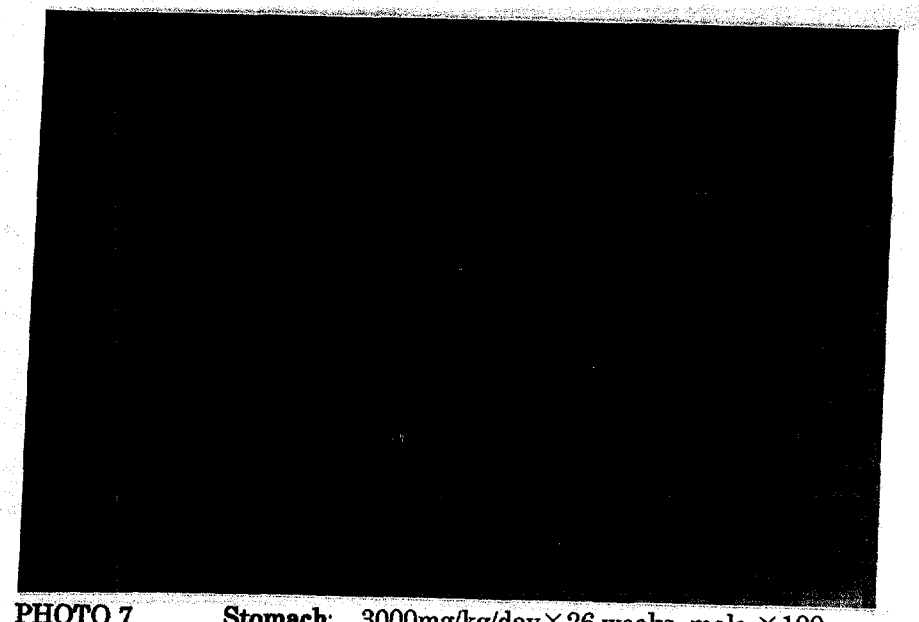


PHOTO 7 **Stomach**: 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

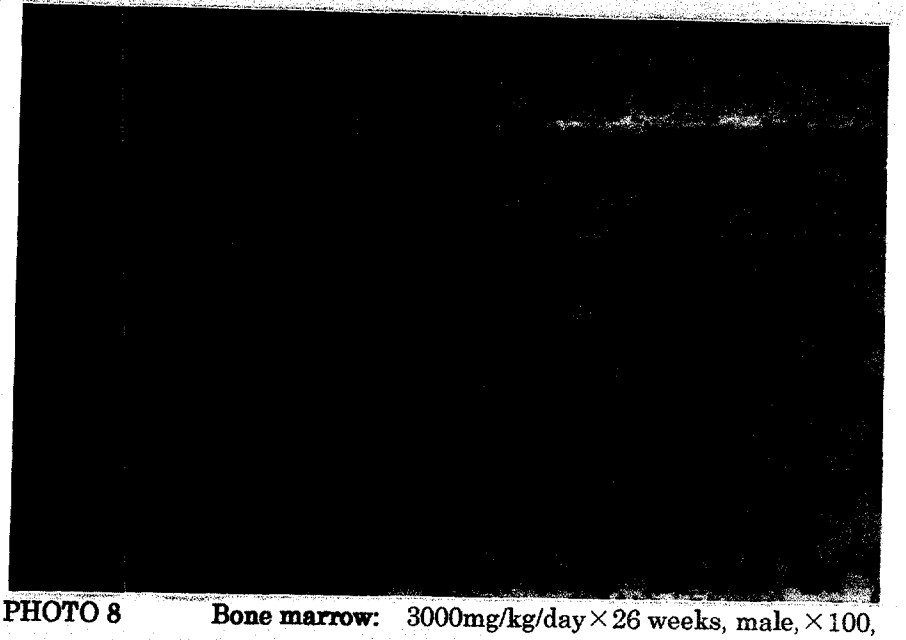


PHOTO 8 **Bone marrow:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

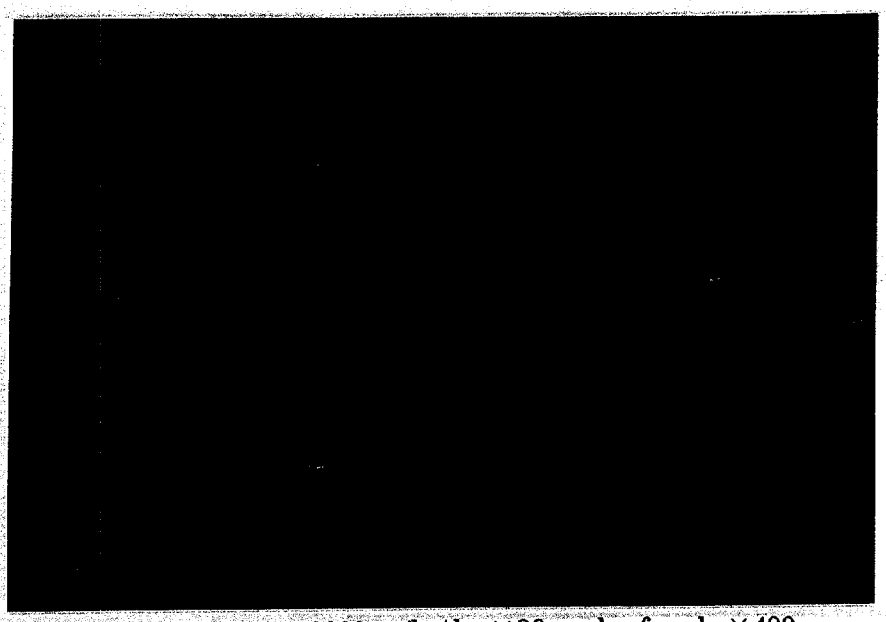


PHOTO 9 **Liver:** 3000mg/kg/day×26 weeks, female, ×400, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

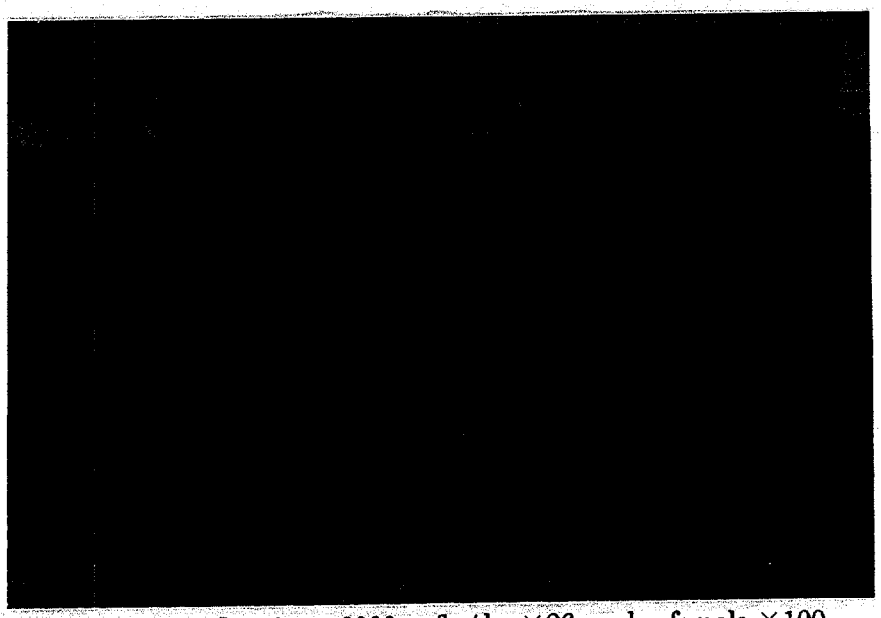


PHOTO 10 **Ovaries:** 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

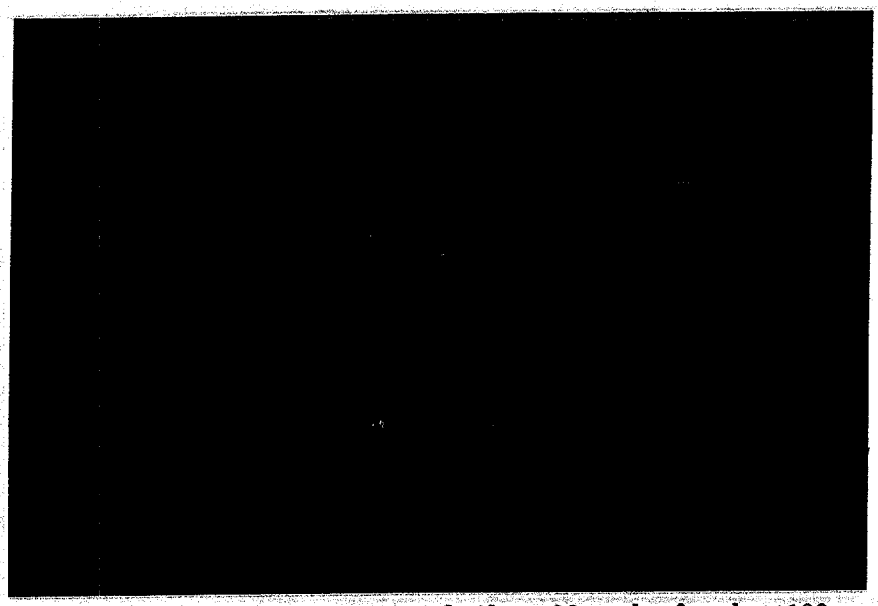


PHOTO 11 **Thyroid**: 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

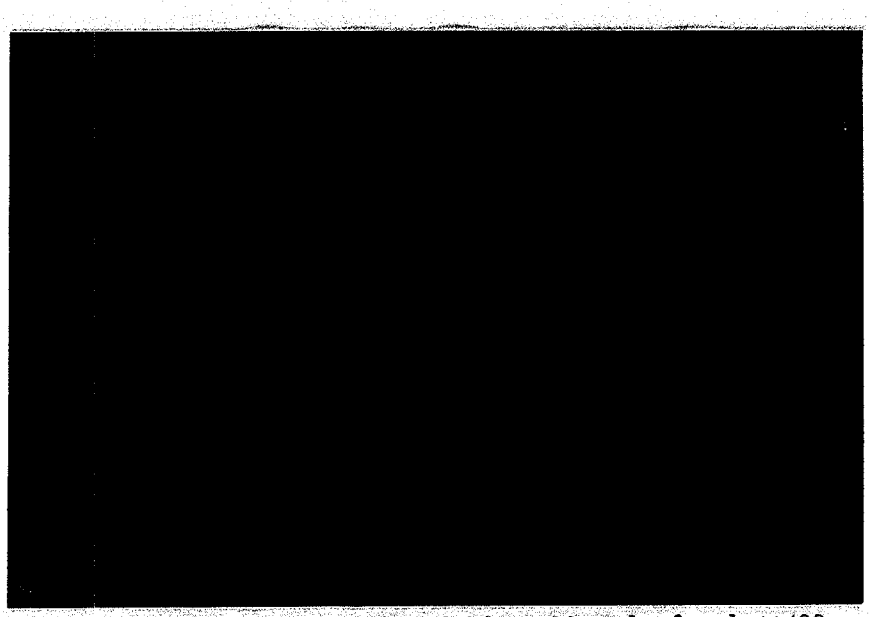


PHOTO 12 **Thyroid**: 3000mg/kg/day×26 weeks, female, ×400, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

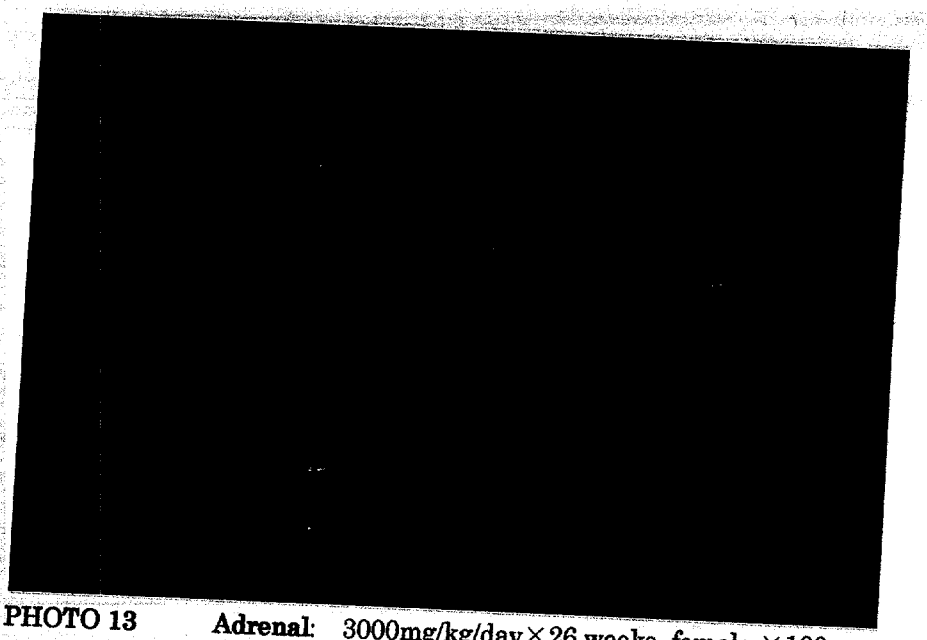


PHOTO 13 **Adrenal**: 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

4.B. II.

Chronic Toxicity Study of Cultured *Agaricus blazei* Murrill (Iwade Strain 101) (Japanease name ; Himematsutake) Preparation, "ABME" Administered Orally in Mice for 26 Weeks.

Hitoshi Ito, M.D., Ph.D. and Keishiro Shimura, M.D.*

Department of Pharmacology Mie University School of Medicine
*Institute of Laboratory Animals, Mie University School of Medicine
2-174, Edobashi, Tsu, Mie 514-0001, Japan

Introduction

A chronic toxicity study of the edible mushroom, *Agaricus blazei* Murrill (Japanese name; Himematsutake) preparation, "ABME" - *Agaricus blazei* Murrill Extract, Japanese name: Himematsutake, was carried out with ICR-Slc strain mice (specific pathogen free animals). The ABME was administered orally for 26 weeks in doses of 0, 500 and 3000 mg/kg/day.

Based on the series of animal experiments studied for the antitumor effect of ABME, the usual dose for human is estimated 25mg/kg. The chronic toxicity study on mice in this report includes 500mg/kg - 20 times and 3000mg/kg - 120 times more dose compared to the usual dose for human.

With the limitation of the capacity of mice stomach and the physical condition of ABME in mind, over 3000mg/kg dose to a mouse would be impossible.

ABME was provided by Iwade Research Institute of Mycology, Japan.

Chronic toxicity studies

Animals were employed 5 weeks old ICR-Slc strain mice, both male and female (Japan SLC, Inc.). The animals were housed and fed in an animal room of the temperature of 23±2℃ and the humidity of 55±5%. Each animal was given solid diet (CLEA Japan CE-2) and water ad libitum.

One group of animals was used of 10 males and 10 females. Doses of administration were determined by the results of subacute toxicity studies, and two grades were adopted; 500 and 3000 mg/kg/day (The maximum dose are able to the oral administration).

Test materials are easily soluble in water but high concentration used the state of suspensions. Their water solutions were, therefore, prepared as to be at a level of 0.1 to 0.15ml per 10g of mice body weight. They were abstained from food for several hours before administration. They were compulsorily administered with a gastric catheter of teflon into the stomach. After the test materials were administered, general symptoms of animals were observed for every day.

Results

(1) Behavior

In mouse administered orally with 500 and 3000 mg/kg for 26 weeks, any abnormal findings that seemed to be caused by the administration of the test material were not observed.

(2) Body Weight Changes (Table 1 and Table 2)

The animals were weighed weekly. No inhibition of body weight gain was found during the periods of the experiments among the test animals, both male and female.

(3) Amount of Diet Ingested (Table 3 and Table 4)

As shown in Table 3 and Table 4, differences between the two administered groups were not observed. As the correct amount of ingested diet was not found, it was impossible to calculate the diet efficacy.

(4) Findings in Hematological Examinations (Table 5, 6, 7 and Table 8)

Hematological examination after 13 weeks of administration was performed on 5 cases of each group, and other items of examinations after 26 weeks of administration were done on 10 cases. No variation of significance was found in red blood cell count, hematocrit value, hemoglobin content, platelet value and white blood cell count. Differential leukocyte was found by fixing blood smear and staining by May-Grünwald Giemsa method. In differential leukocyte count, no abnormal findings were found due to the administration of the test material.

(5) Biochemical Examination of Blood (Table 9, 10, 11 and Table 12)

Biochemical examinations of blood after 13 weeks of administration were performed on 5 cases each of the groups. With regard to glucose, urea, total protein, albumin, alkaline phosphatase, GOT, GPT and total cholesterol value, no abnormal data was found in the female animals of the 500 mg/kg and 3000 mg/kg groups at 13^{th} and 26^{th} week.

(6) Findings in Urine (Table 13 and Table 14)

Urine protein was assayed in the concentration of trace to 100mg/dl in most of the groups, regardless of the administered or the control. Inspecting , pH, urobilinogen, bilirubin, ketone body and glucose, no abnormal data was found in all groups.

(7) Findings at Autopsy Organ Weight (Table 15, 16, 17 and Table 18)

At the end of administration, blood was sampled under anesthesia of ether. Immediately after the mice were sacrificed by blood-letting, autopsy was performed, organs were excised, principal organs were weighted wet, fixed with 10% formalin, and put to the histopathological examinations. As shown in Table 15, 16, 17 and Table 18, no remarkable change was found between the control group and the treated groups in either absolute organ weight or mean comparative organ weight. The changes found were not considered to be caused by the test material.

(8) Histopathological Findings (Table 19, 20, 21 and Table 22, PHOTO 1—PHOTO 14)

After autopsy and gross observation of changes, the organs were fixed with 10% formalin, embedded in paraffin and cut in slices ca. 6 μ thick, then stained with hematoxylin and eosine.

Microscopic examinations were performed on 5 samples each of the groups at the end of 13 weeks and 26weeks after the administration. Histopathological examination was conducted by Sensake Naruse, M.D., at Department of Pathology, Mie University School of Medicine, Tsu, Mie, 514-0001, Japan.

Findings in the survived mice are as follows:

Lungs : Two cases including the control group at 26 weeks showed a slight chronic bronchitis and inflammation of interstitial cells.

Liver : Almost no difference between the control and the administered groups; a slight cell infiltration and degeneration of liver were observed in 1 case of the control group.

Kidneys : In one case of the 3000mg/kg group, kidney tubules were found enlarged like cyst, and may have been caused by pyelitis and nephritis. However, a slight pyelitis and nephritis were found in the control group, too.

Spleen : A slight hemosiderosis was observed in a few case including those of the control and the administered groups.

* No marked changes were observed in brain, heart, testes, ovaries, thymus, pituitary, adrenals, pancreas and digestive tracts.

Summary

A chronic toxicity of edible mushroom, *Agaricus blazei* Murrill (Japanese name: Himematsutake) preparation, "ABME" was studied with ICR strain mice.

ABME was administered orally for 26 weeks in dose of 0 (control), 500 and 3000 mg/kg/day. During the period of oral administration for 26 weeks, no general symptoms to be marked were observed in ICR strain mice, and there was no specific change of male and female mice.

With regard to the amount of diet ingested, no significant change was found in all the administered group.

No inhibition of body weight gain was found during the periods of the experiments among the test animals, both male and female.

In hematological findings, any significant variation in red blood cell count, hematocrit value, hemoglobin content, platelet value and white blood cell count was not found. In differential count, too, no abnormal findings due to the administration of the test material was found.

In biochemical examination of blood, no change was found in glucose, urea, total protein, albumin, alkaline phosphatase, GOT, GPT and total cholesterol value.

No abnormality was found in the pH, urobilinogen, bilirubin, ketone body, protein and glucose in the control and the administered groups.

In assaying organ weight, no change was found in either the administered group of male or female mice compared with the control group.

In the histopathogical examinations, any abnormal figures specific to the administered group compared with the control group was not observed in mice. Furthermore, any toxicity to be caused by ABME could not be found.

As a conclusion of chronic toxicity studies in mice, the safety dose for mice was estimated to be over 3000 mg/kg/day, but the sure intoxication dose could not be determined.

End of report

Table 1 Body weight changes in male mice given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Male (g)			
	Number of mice	Control	500	3000
0	15	28.4	28.1	28.7
1	15	32.7	31.8	33.4
2	15	34.5	35.3	36.2
3	15	36.2	37.0	36.8
4	15	38.0	39.2	37.8
5	15	39.9	40.8	39.4
6	15	41.3	42.0	41.0
7	15	42.4	43.5	41.9
8	15	43.9	45.0	42.7
9	15	44.5	45.5	43.2
10	15	45.5	46.7	44.1
11	15	45.3	47.2	45.4
12	15	45.9	47.0	45.2
13	15	46.4	48.3	46.7
14	10	46.3	49.0	47.2
15	10	46.4	49.4	47.7
16	10	47.0	50.6	47.9
17	10	47.8	51.3	47.6
18	10	48.3	51.7	48.5
19	10	48.9	52.0	48.7
20	10	49.8	52.7	49.2
21	10	51.0	53.4	51.6
22	10	51.9	53.7	52.1
23	10	52.2	53.5	53.0
24	10	52.7	54.1	53.4
25	10	53.0	54.2	53.7
26	10	53.1	54.5	53.9

Table 2 , Body weight changes in female mice given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Female (g)			
	Number of mice	Control	500	3000
0	15	22.8	23.4	23.4
1	15	25.5	26.0	26.6
2	15	26.3	26.9	27.9
3	15	28.2	29.0	28.9
4	15	29.7	30.5	30.5
5	15	30.3	32.0	31.9
6	15	31.0	32.2	32.3
7	15	32.2	33.2	33.5
8	15	34.0	34.4	34.7
9	15	34.4	35.2	35.3
10	15	35.6	35.5	36.3
11	15	35.5	36.6	37.7
12	15	35.7	37.5	39.3
13	15	36.6	38.0	39.4
14	10	36.5	38.7	41.3
15	10	37.0	39.0	41.5
16	10	37.8	39.8	42.4
17	10	38.4	41.0	43.0
18	10	39.9	41.7	43.9
19	10	41.6	42.5	44.5
20	10	41.8	43.6	45.6
21	10	42.7	44.0	45.6
22	10	43.4	44.8	46.0
23	10	43.4	44.7	45.9
24	10	44.0	45.1	46.1
25	10	44.5	45.3	46.2
26	10	44.8	45.9	46.3

Table 3' Food consumption of male mice given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Male (g)			
	Number of mice	Control	500	3000
1	15	4.9±0.3	5.2±0.4	5.2±0.4
2	15	5.6±0.3	5.5±0.3	5.9±0.5
3	15	7.5±0.8	7.8±0.7	6.5±0.6
4	15	8.2±0.9	9.3±0.9	8.1±0.8
5	15	8.7±0.7	9.0±0.6	8.1±0.5
6	15	8.8±0.4	10.1±0.5	9.0±0.6
7	15	9.3±0.5	10.1±0.4	9.4±0.7
8	15	8.9±0.5	9.6±0.6	8.5±0.6
9	15	8.1±0.7	9.7±0.5	8.3±0.3
10	15	8.6±0.5	9.8±0.5	8.9±0.4
11	15	8.8±0.8	9.4±0.6	9.3±0.7
12	15	8.6±0.7	9.5±0.5	9.2±0.5
13	15	8.7±0.6	9/8±0.7	9.5±0.8
14	10	8.4±0.5	9.6±0.8	8.3±0.7
15	10	8.4±0.9	9.5±0.6	8.6±0.6
16	10	8.3±0.5	9.5±0.7	8.7±0.7
17	10	7.9±0.8	9.6±0.8	9.0±0.9
18	10	8.6±0.7	10.1±1.0	9.1±0.8
19	10	8.2±0.6	9.7±0.8	8.6±0.7
20	10	8.5±0.8	9.7±0.9	8.7±0.6
21	10	8.5±0.8	9.8±0.7	8.8±0.6
22	10	8.7±0.9	9.7±0.9	8.9±0.7
23	10	8.8±0.7	8.5±0.5	9.1±0.5
24	10	8.4±0.6	8.5±0.6	9.0±0.7
25	10	8.7±0.7	9.0±0.4	9.1±0.5
26	10	8.9±0.5	9.1±0.5	9.0±0.4

Values represent mean ± standard error (g/day/mouse)

Table 4 Food consumption of female mice given ABME orally for 26 weeks

Dosing periods (weeks)	Dose level (mg/kg/day)			
	Female (g)			
	Number of mice	Control	500	3000
1	15	4.3±0.2	4.6±0.3	4.2±0.2
2	15	4.8±0.4	4.9±0.3	4.1±0.3
3	15	4.9±0.3	4.9±0.2	4.7±0.2
4	15	5.3±0.4	5.2±0.3	5.2±0.2
5	15	5.6±0.3	6.1±0.3	5.3±0.4
6	15	5.6±0.3	6.1±0.3	5.9±0.3
7	15	6.0±0.2	6.1±0.3	6.5±0.4
8	15	6.2±0.7	7.2±0.8	6.3±0.5
9	15	6.1±0.5	7.2±0.7	7.0±0.6
10	15	6.3±0.7	7.3±0.8	7.2±0.7
11	15	6.8±0.5	7.9±0.9	6.9±0.7
12	15	6.0±0.4	6.8±0.5	7.1±0.7
13	15	6.4±0.5	7.0±0.7	7.1±0.6
14	10	6.3±0.3	7.2±0.6	6.7±0.4
15	10	6.2±0.5	7.5±0.5	6.9±0.4
16	10	6.7±0.7	6.9±0.6	7.2±0.6
17	10	6.5±0.5	7.0±0.7	6.7±0.6
18	10	6.9±0.9	7.1±0.5	6.8±0.5
19	10	6.4±0.6	7.1±0.5	7.0±0.7
20	10	6.7±0.5	6.8±0.5	7.0±0.4
21	10	6.8±0.7	7.4±0.6	6.7±0.6
22	10	6.6±0.5	7.2±0.7	6.8±0.6
23	10	6.7±0.4	7.0±0.8	7.1±0.5
24	10	6.7±0.3	7.3±0.8	6.6±0.5
25	10	6.9±0.6	7.4±0.5	7.1±0.8
26	10	7.1±0.7	7.6±0.5	7.2±0.7

Values represent mean ± standard error (g/day/mouse)

Table 5 Hematological findings in male mice given ABME orally for 13 weeks

Male											
Dose level (mg/kg/day)	Number of mice	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%)				
							L	M	N	E	B
Control	5	778±22	46.8±1.8	15.3±0.3	120±14.7	51±9.4	71.9±2.7	1.0±0.4	26.3±2.6	0.8±0.2	0
500	5	777±20	47.1±1.7	15.0±0.6	115±13.0	50±7.3	70.6±2.5	1.4±0.3	27.2±2.4	0.8±0.1	0
3000	5	780±34	46.1±1.3	15.6±0.3	118±14.1	48±5.7	72.8±2.5	1.0±0.4	25.5±2.5	0.7±0.2	0

Values represent mean ± standard error
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 6 Hematological findings in female mice given ABME orally for 13 weeks

Female											
Dose level (mg/kg/day)	Number of mice	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%)				
							L	M	N	E	B
Control	5	766±33	46.7±1.1	14.9±0.2	117±9.2	49±3.3	76.3±1.6	0.8±0.3	21.5±1.8	1.4±0.3	0
500	5	785±19	47.1±1.0	14.8±0.8	115±7.0	51±3.8	75.2±1.8	1.2±0.2	22.4±2.8	1.2±0.2	0
3000	5	780±28	47.2±1.2	15.1±0.7	123±9.7	46±2.2	76.2±2.8	1.4±0.3	21.5±2.5	0.9±0.3	0

Values represent mean ± standard error
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 8 Hematological findings in female mice given ABME orally for 26 weeks

Female											
Dose level (mg/kg/day)	Number of mice	RBC ($\times 10^4/mm^3$)	Ht (%)	Hb (g/dl)	BP ($\times 10^4/mm^3$)	WBC ($\times 10^2/mm^3$)	Differential count (%)				
							L	M	N	E	B
Control	10	883±52	44.7±1.3	14.7±0.6	129±7.6	42±4.3	71.9±3.6	1.3±0.4	26.0±3.0	0.8±0.3	0
500	10	920±73	45.1±2.6	15.2±0.7	130±5.3	39±2.3	70.9±2.8	1.1±0.3	27.0±3.2	1.0±0.3	0
3000	10	914±46	44.0±2.1	14.9±0.6	126±3.9	44±3.5	67.4±3.5	1.9±0.5	30.0±3.3	0.7±0.4	0

Values represent mean ± standard error
RBC (Red blood cell) : TOA Microcell Counter CC-108
Ht (Hematocrit) : Microhematocrit method
Hb (Hemoglobin) : TOA Hemoglobin Counter Hb-100
BP (Blood platelet) : TOA Platelet Counter PL-100
WBC (White blood cell) : TOA Microcell Counter CC-108
L (Lymphocyte), M (Monocyte), N (Neutrophil), E (Eosinocyte) and B (Basocyte) :
Leucocyte ratio (May-Grunwald Giemsa stained method)

Table 9 Biochemical findings in male mice given ABME orally for 13 weeks

Male									
Dose level (mg/kg/day)	Number of mice	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)
Control	5	127±29	25±4.1	4.6±0.3	1.5±0.3	175±29.3	52±9.7	22±4.3	127±26
500	5	132±30	27±3.7	4.2±0.5	1.4±0.2	198±32.4	54±7.3	26±5.1	130±32
3000	5	116±21	22±3.5	5.0±0.4	1.5±0.3	170±31.9	59±9.8	23±3.4	121±23

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method

Table 10 Biochemical findings in female mice given ABME orally for 13 weeks

Female									
Dose level (mg/kg/day)	Number of mice	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)
Control	5	133±31	24±2.7	4.6±0.1	1.4±0.2	241±53.3	60±9.8	24±5.1	136±18
500	5	140±29	21±3.4	4.4±0.2	1.3±0.1	221±35.1	57±6.7	21±4.7	120±15
3000	5	128±26	20±2.1	4.5±0.3	1.4±0.1	199±41.4	65±9.7	27±5.6	109±17

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method

Table 11 Biochemical findings in male mice given ABME orally for 26 weeks

Male									
Dose level (mg/kg/day)	Number of mice	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)
Control	10	102±6.3	22±3.9	4.8±0.3	1.4±0.2	160±22.6	76±10.2	27±5.3	119±23
500	10	96±5.1	21±2.4	4.5±0.2	1.6±0.3	154±20.3	72±9.7	25±4.1	107±15
3000	10	98±5.2	18±2.0	4.9±0.2	1.5±0.2	157±16.5	68±8.1	28±3.6	98±19

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method

Table 12 Biochemical findings in female mice given ABME orally for 26 weeks

Female									
Dose level (mg/kg/day)	Number of mice	Glucose (mg/dl)	Urea nitrogen (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Alkaline phoshatase (IU/l)	GOT (IU/l)	GPT (IU/l)	Total cholesterol (mg/dl)
Control	10	93±19	19±2.7	5.1±0.4	1.5±0.2	159±36.0	97±21	37±4.9	110±23
500	10	90±12	22±3.4	4.9±0.3	1.5±0.1	162±41.4	91±34	35±5.3	98±16
3000	10	89±11	20±2.1	5.3±0.3	1.4±0.1	158±29.6	109±42	32±3.7	101±18

Values represent mean ± standard error
* Significantly different from control at $p < 0.05$
Glucose : GLK/G6PDH method
Urea nitrogen : Urease/GLDH method
Albumin : BCG method
Alkaline phosphatase : King-Armstrong method
GOT : NADH-UV (IFCC method)
GPT : NADH-UV (IFCC method)
Total cholesterol : CE/CO/POD method

Table 13 Urinalysis of male mice given ABME orally for 26 weeks

Male												
Dosing period (week)	Dose level (mg/kg/day)	Number of mice	Appearance	pH			Occult blood	Urobilinogen (Ehrlich unit/dl)	Bilirubin (0.4–0.8 mg/dl)	Ketone body	Protein	Glucose
				6	7	8						
13	Control	10	Normal	4	1	0	−	0.1–1	−	−	±～+	−
	500	10	Normal	3	2	0	−	0.1	−	−	−～±	−
	3000	10	Normal	4	1	0	−	0.1	−	−	±～+	−
26	Control	10	Normal	5	0	0	−	0.1–1	−	−	±～+	−
	500	10	Normal	5	0	0	−	0.1	−	−	−～±	−
	3000	10	Normal	4	1	0	−	0.1–1	−	−	−～±	−

pH : pH meter,
Occult blood, Urobilinogen, Bilirubin, Ketone body,
Protein and Glucose : Uro-Labstix (Ames reagent strips for urinalysis)

Table 14 Urinalysis of female mice given ABME orally for 26 weeks

Female													
Dosing period (week)	Dose level (mg/kg/day)	Number of mice	Appearance	pH			Occult blood	Urobilinogen (Ehrlich unit/dl)	Bilirubin (0.4–0.8 mg/dl)	Ketone body	Protein	Glucose
				6	7	8						
13	Control	10	Normal	3	2	0	−	0.1–1	−	−	±～+	−
	500	10	Normal	3	2	0	−	0.1–1	−	−	±～+	−
	3000	10	Normal	4	1	0	−	0.1	−	−	−～±	−
26	Control	10	Normal	4	1	0	−	0.1–1	−	−	±～+	−
	500	10	Normal	5	0	0	−	0.1		−	−～±	−
	3000	10	Normal	4	1	0	−	0.1–1	−	−	±～+	−

pH : pH meter,
Occult blood, Urobilinogen, Bilirubin, Ketone body,
Protein and Glucose : Uro-Labstix (Ames reagent strips for urinalysis)

Table 15 Organ weights in male mice given ABME orally for 13 weeks

Male												
Dose level (mg/kg/day)	Number of mice	Final body wt. (g)	Brain (mg)	Heart (mg)	Lung (mg)	Liver (g)	Kidneys (mg)	Spleen (mg)	Testes (mg)	Thymus (mg)	Pituitary (mg)	Adrenals (mg)
Control	5	46.4±2.7	543±14 (1.170)	163±8 (0.351)	181±15 (0.390)	1.70±0.21 (3.664)	460±29 (0.991)	122±12 (0.263)	344±25 (0.741)	55.8±4.8 (0.120)	2.8±0.2 (0.006)	23.7±3.4 (0.051)
500	5	48.3±3.1	552±17 (1.143)	166±7 (0.344)	187±21 (0.387)	1.55±0.23 (3.209)	477±31 (0.988)	124±23 (0.257)	345±29 (0.714)	51.4±3.2 (0.106)	2.9±0.1 (0.006)	29.1±4.0 (0.060)
3000	5	46.7±2.0	549±15 (1.176)	167±9 (0.358)	173±10 (0.370)	1.84±0.30 (3.940)	462±30 (0.989)	130±9 (0.278)	359±24 (0.769)	54.0±2.2 (0.116)	3.0±0.2 (0.006)	24.5±7.0 (0.052)

Values represent mean ± standard error.
Values in parentheses represent mean comparative organ weights in grams or milligrames per 100g body weight.

Table 16 Organ weights in female mice given ABME orally for 13 weeks

Female												
Dose level (mg/kg/day)	Number of mice	Final body wt. (g)	Brain (mg)	Heart (mg)	Lung (mg)	Liver (g)	Kidneys (mg)	Spleen (mg)	Ovaries (mg)	Thymus (mg)	Pituitary (mg)	Adrenals (mg)
Control	5	36.6±2.0	545±16	152±8	190±11	1.67±0.09	369±18	134±16	13.3±0.9	56.4±5.1	3.1±0.3	12.4±1.3
			(1.489)	(0.415)	(0.519)	(4.563)	(1.008)	(0.366)	(0.036)	(0.154)	(0.008)	(0.034)
500	5	38.0±2.5	557±12	158±4	197±7	1.70±0.21	377±22	146±17	13.5±1.4	59.8±4.0	3.3±0.4	12.6±3.1
			(1.466)	(0.416)	(0.518)	(4.474)	(0.992)	(0.384)	(0.036)	(0.157)	(0.009)	(0.033)
3000	5	39.4±3.1	570±10	159±5	199±13	1.88±0.20	380±21	149±15	14.8±1.8	59.9±7.4	3.5±0.2	13.4±4.5
			(1.447)	(0.404)	(0.505)	(4.772)	(0.964)	(0.378)	(0.038)	(0.152)	(0.009)	(0.034)

Values represent mean ± standard error.
Values in parentheses represent mean comparative organ weights in grams or milligrames per 100g body weight.

Table 17 Organ weights in male mice given ABME orally for 26 weeks

Male												
Dose level (mg/kg/day)	Number of mice	Final body wt. (g)	Brain (mg)	Heart (mg)	Lung (mg)	Liver (g)	Kidneys (mg)	Spleen (mg)	Testes (mg)	Thymus (mg)	Pituitary (mg)	Adrenals (mg)
Control	10	53.1±4.2	564±18 (1.062)	233±12 (0.439)	231±15 (0.435)	2.48±0.24 (4.670)	709±28 (1.335)	134±12 (0.252)	544±15 (1.024)	45.7±3.9 (0.086)	3.0±0.2 (0.006)	26.8±3.5 (0.050)
500	10	54.5±4.9	566±19 (1.039)	246±10 (0.451)	247±20 (0.453)	2.52±0.18 (4.624)	712±30 (1.306)	140±26 (0.257)	544±9 (0.996)	45.4±2.3 (0.083)	3.2±0.2 (0.006)	26.1±2.7 (0.048)
3000	10	53.9±4.3	577±13 (1.071)	239±6 (0.443)	243±17 (0.451)	2.53±0.09 (4.694)	730±29 (1.354)	142±11 (0.263)	553±14 (1.026)	44.0±1.7 (0.082)	3.4±0.2 (0.006)	27.3±3.3 (0.051)

Values represent mean ± standard error.
Values in parentheses represent mean comparative organ weights in grams or milligrames per 100g body weight.

Table 18 Organ weights in female mice given ABME orally for 26 weeks

Female												
Dose level (mg/kg/day)	Number of mice	Final body wt. (g)	Brain (mg)	Heart (mg)	Lung (mg)	Liver (g)	Kidneys (mg)	Spleen (mg)	Ovaries (mg)	Thymus (mg)	Pituitary (mg)	Adrenals (mg)
Control	10	44.8±5.7	566±13	165±6	218±10	2.02±0.15	417±23	147±13	22.7±0.9	72.1±3.4	2.9±0.2	14.3±2.1
			(1.263)	(0.368)	(0.487)	(4.509)	(0.931)	(0.328)	(0.051)	(0.161)	(0.006)	(0.032)
500	10	45.9±4.9	577±10	163±8	209±9	2.11±0.24	412±24	150±17	23.2±1.7	74.6±4.1	3.1±0.3	14.8±3.1
			(1.257)	(0.355)	(0.455)	(4.597)	(0.898)	(0.327)	(0.051)	(0.163)	(0.007)	(0.032)
3000	10	46.3±5.6	581±15	171±7	214±9	2.23±0.33	446±20	154±12	25.5±2.1	75.8±8.3	2.9±0.1	14.4±1.4
			(1.255)	(0.369)	(0.462)	(4.816)	(0.963)	(0.333)	(0.055)	(0.164)	(0.006)	(0.031)

Values represent mean ± standard error.
Values in parentheses represent mean comparative organ weights in grams or milligrames per 100g body weight.

Table 19 Summary of histopathological findings in male mice received daily oral administration of ABME for 13 weeks

Number of individual animal	Control (0)					ABME-500					ABME-3000				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Liver; nuclear hyperplasia	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	±	−	−	−	−	−	−	−	−	−	−	−	−	−
degeneration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
necrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Kidneys; hyaline droplets	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
fibrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Spleen; hemosiderin	−	−	±	±	−	−	−	−	−	−	−	−	−	−	−
Heart; cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Thymus; involution	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Brain	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Lungs	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Testis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Pituitary	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Adrenals	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Stomach	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−

±; Very slight alteration

Table 20 Summary of histopathological findings in female mice received daily oral administration of ABME for 13 weeks

Number of individual animal	Control (0)					ABME-500					ABME-3000				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Liver; nuclear hyperplasia	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
degeneration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
necrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Kidneys; hyaline droplets	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
fibrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Spleen; hemosiderin	−	±	−	−	−	−	−	−	−	±	−	−	−	−	−
Heart; cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Thymus; involution	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Brain	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Lungs	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Ovaries	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Pituitary	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Adrenals	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Stomach	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−

±; Very slight alteration

Table 21 Summary of histopathological findings in male mice received daily oral administration of ABME for 26 weeks

Number of individual animal	Control (0)					ABME-500					ABME-3000				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Liver; nuclear hyperplasia	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
degeneration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
necrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Kidneys; hyaline droplets	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
fibrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Spleen; hemosiderin	±	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Heart; cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Thymus; involution	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Brain	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Lungs	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Testis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Pituitary	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Adrenals	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Stomach	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−

±; Very slight alteration

Table 22 Summary of histopathological findings in female mice received daily oral administration of ABME for 26 weeks

Number of individual animal	Control (0)					ABME-500					ABME-3000				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Liver; nuclear hyperplasia	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
degeneration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
necrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Kidneys; hyaline droplets	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
fibrosis	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Spleen; hemosiderin	−	±	−	−	−	−	−	−	−	−	−	−	±	−	−
Heart; cell infiltration	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Thymus; involution	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Brain	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Lungs	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Ovaries	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Pituitary	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Adrenals	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Stomach	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−

±; Very slight alteration

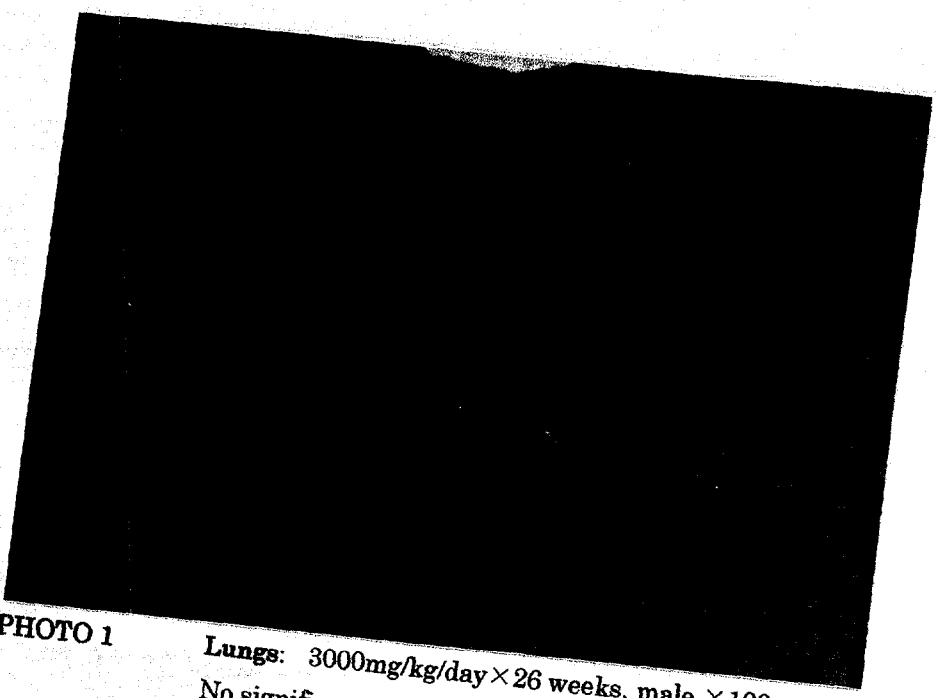


PHOTO 1 **Lungs:** 3000mg/kg/day × 26 weeks, male, × 100, No significant change (Not different from treated group of 3000mg/kg/day × 13 weeks and control group)

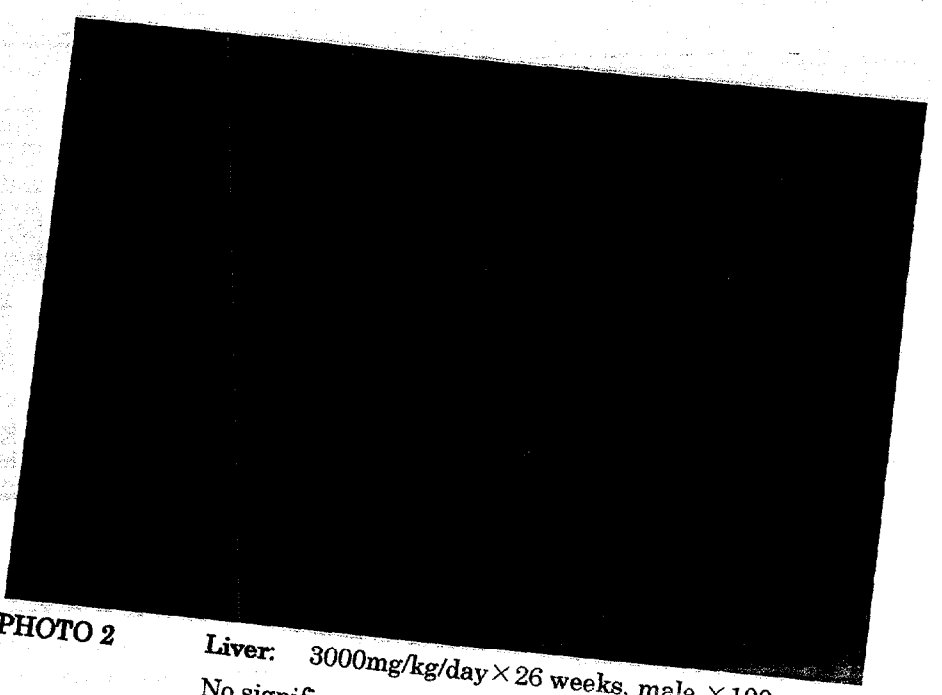


PHOTO 2 **Liver:** 3000mg/kg/day × 26 weeks, male, × 100, No significant change (Not different from treated group of 3000mg/kg/day × 13 weeks and control group)

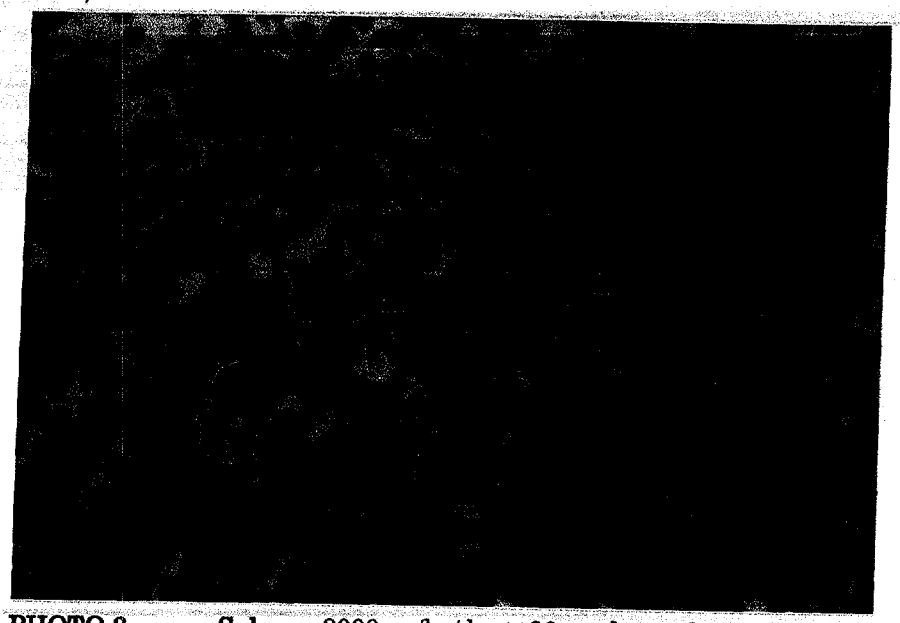


PHOTO 3 **Spleen**: 3000mg/kg/day×26 weeks, male, ×400,
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

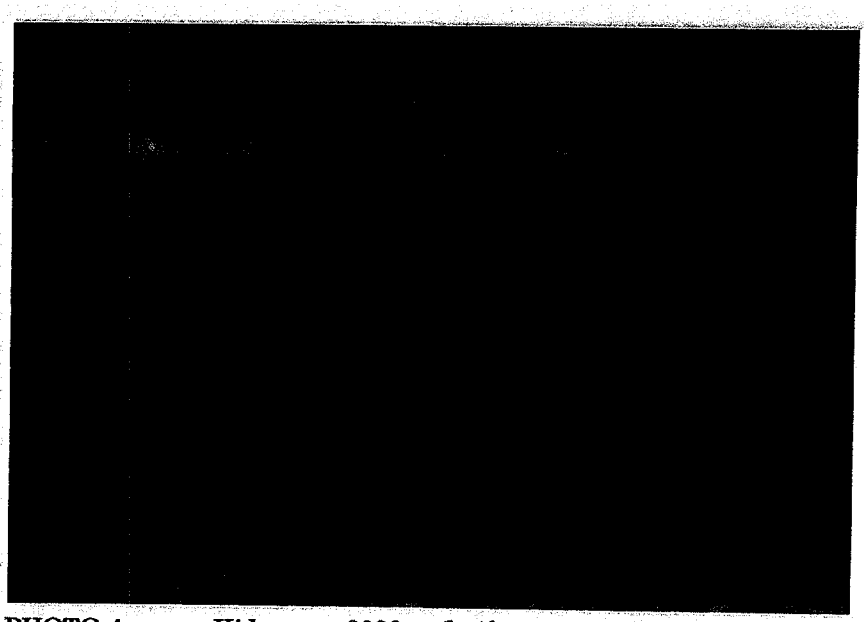


PHOTO 4 **Kidneys**: 3000mg/kg/day×26 weeks, male, ×100,
No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

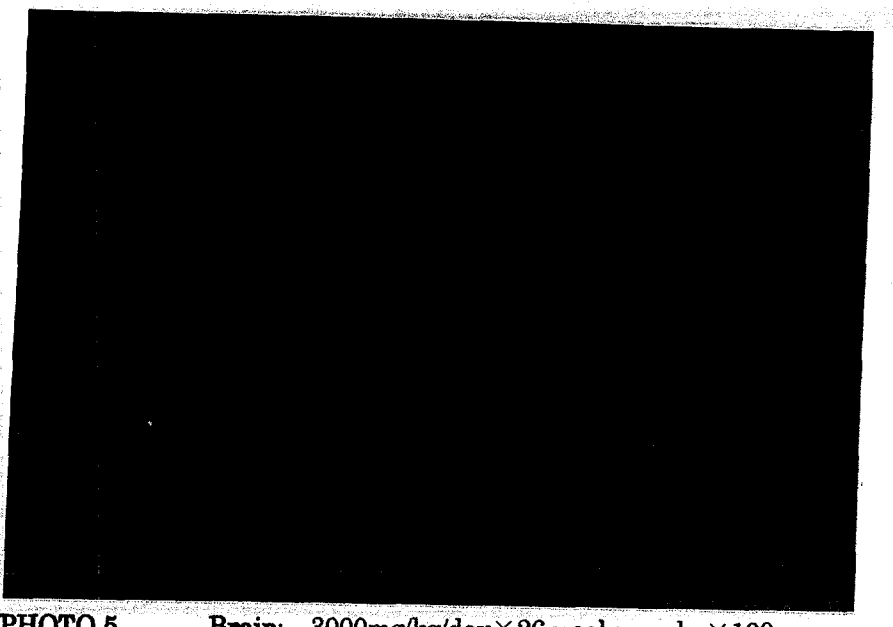


PHOTO 5 **Brain**: 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

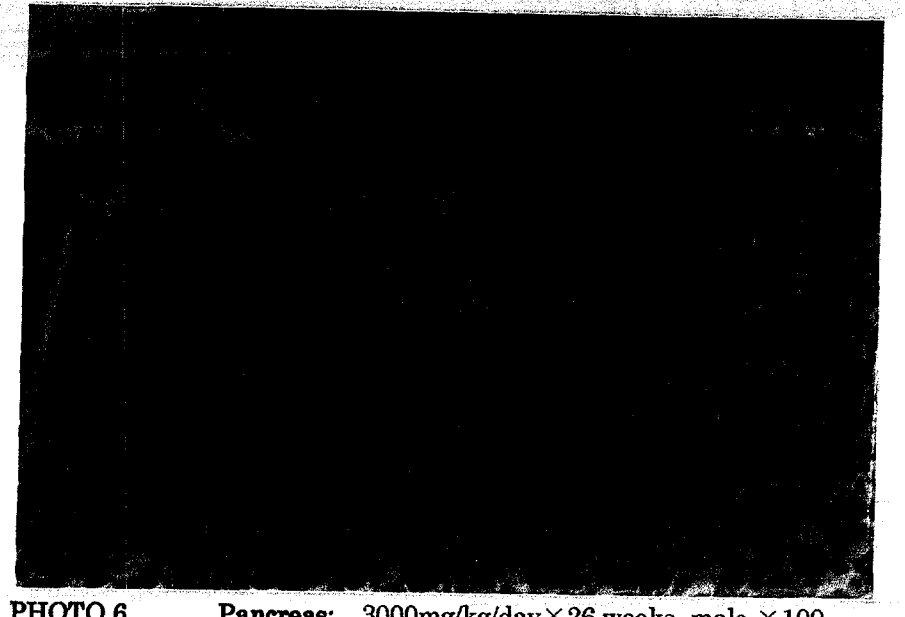


PHOTO 6 **Pancreas:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

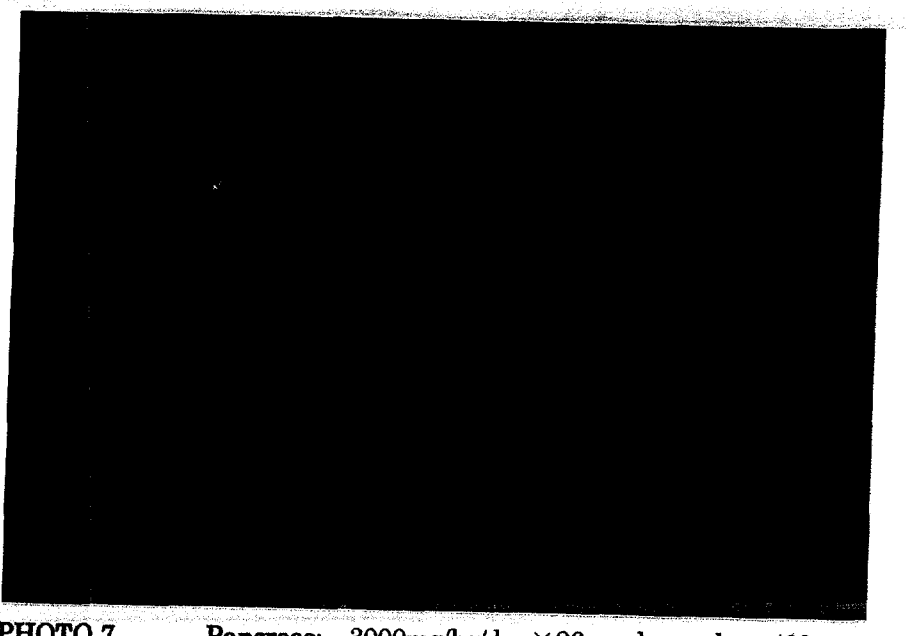


PHOTO 7 **Pancreas:** 3000mg/kg/day×26 weeks, male, ×400, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

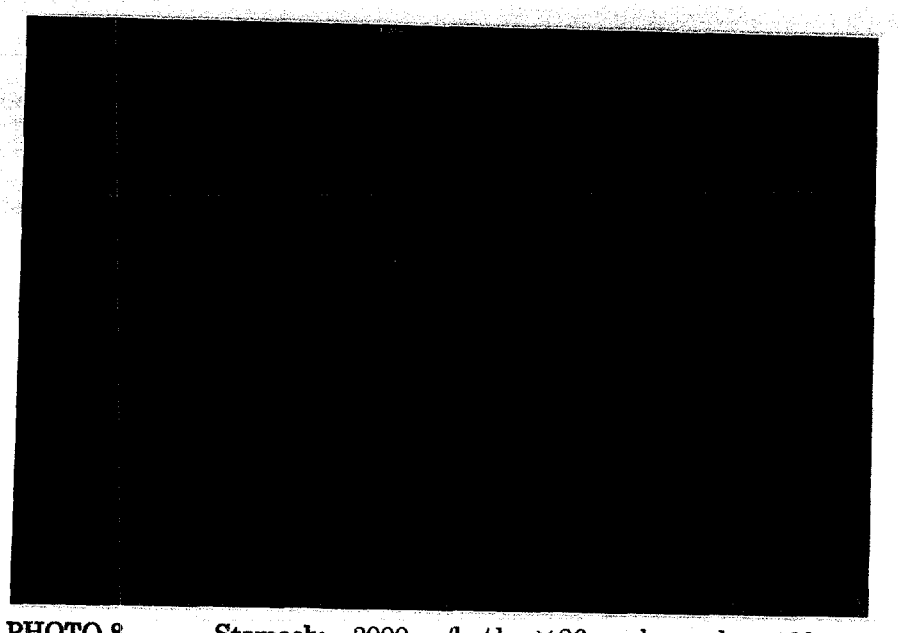


PHOTO 8 **Stomach:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

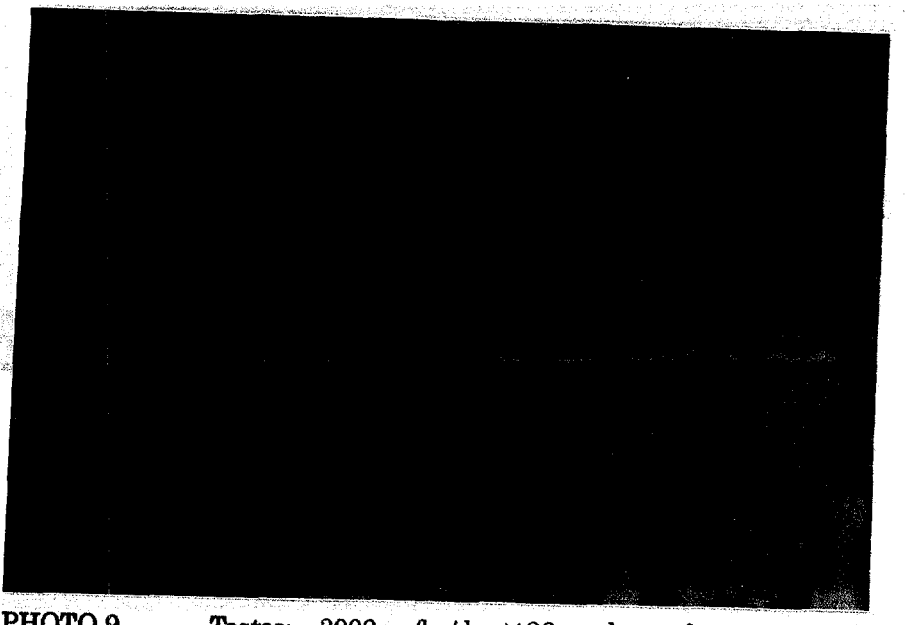


PHOTO 9 **Testes:** 3000mg/kg/day×26 weeks, male, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

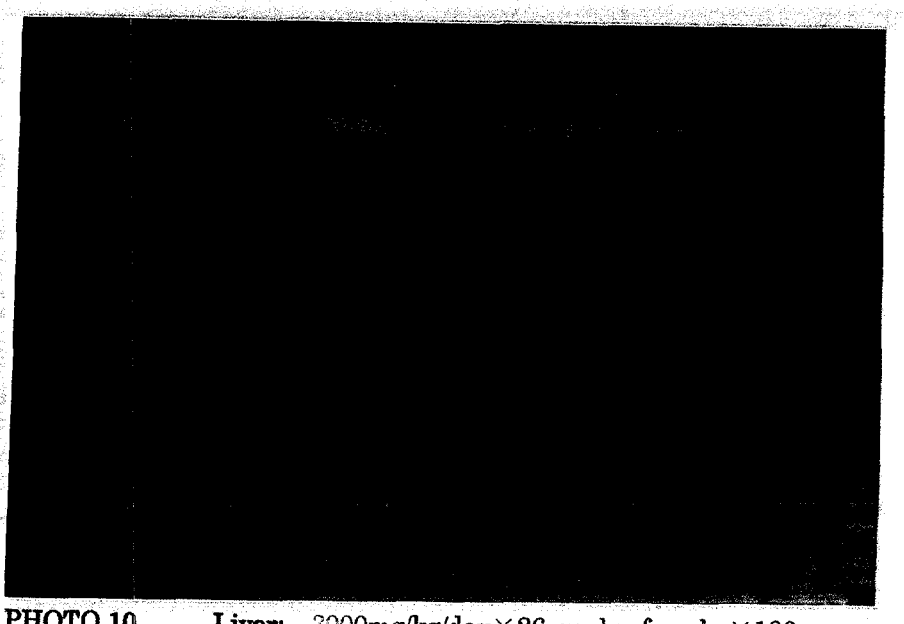


PHOTO 10 **Liver:** 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

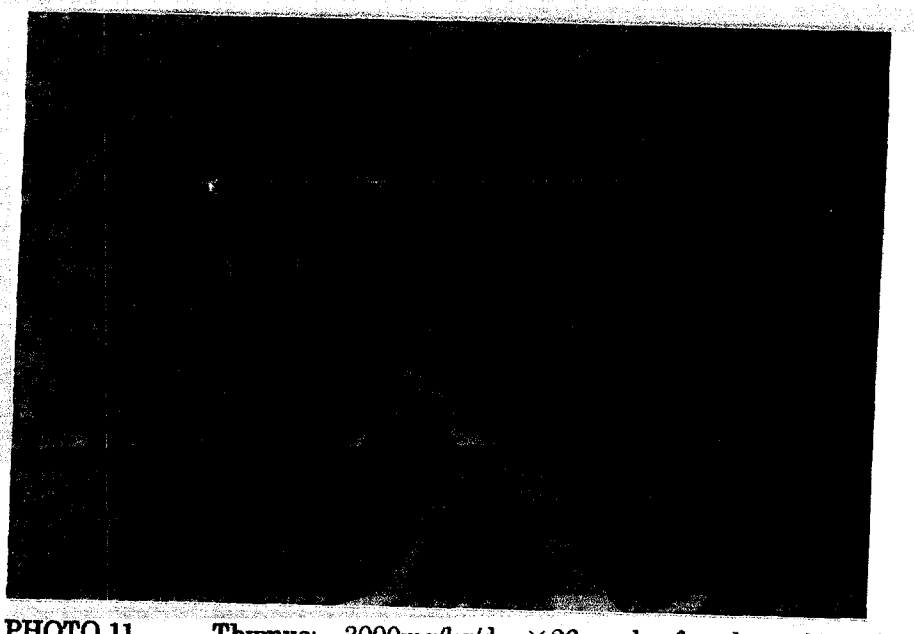


PHOTO 11 **Thymus**: 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

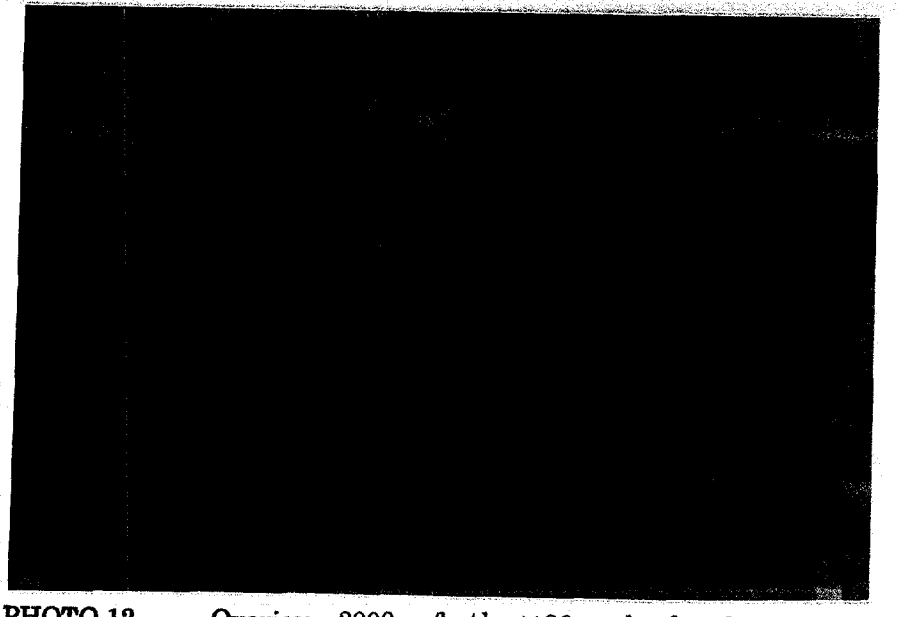


PHOTO 12 **Ovaries**: 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

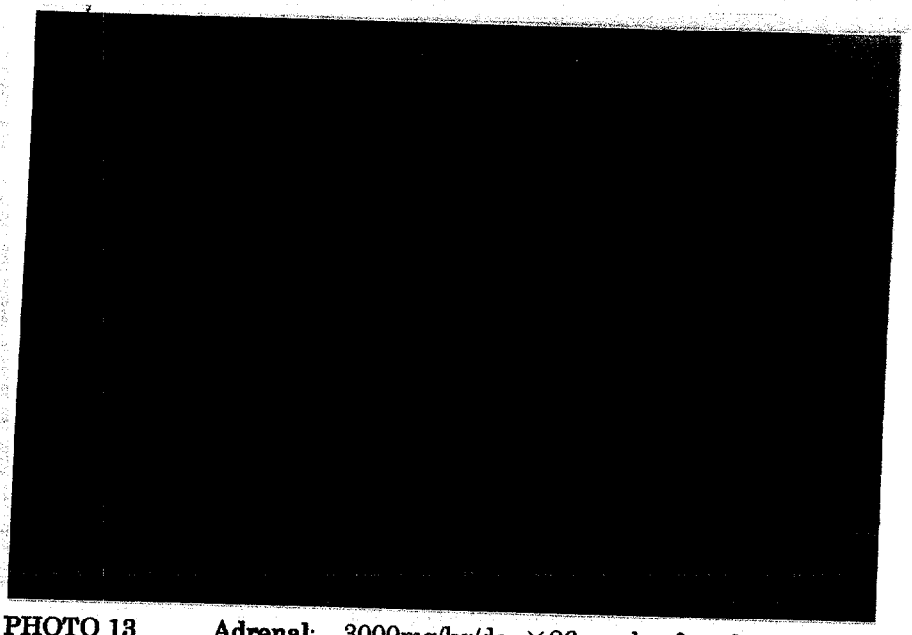


PHOTO 13 **Adrenal**: 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)

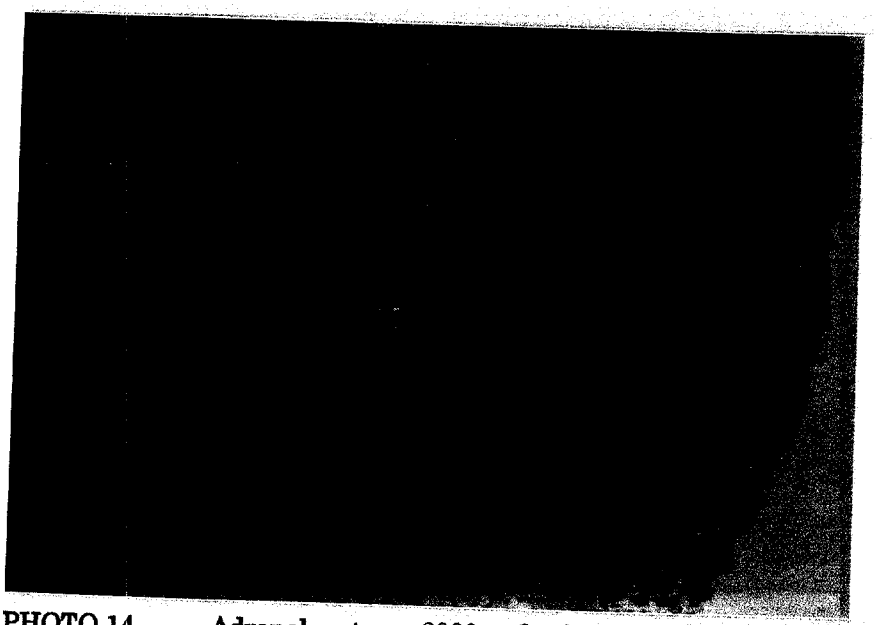
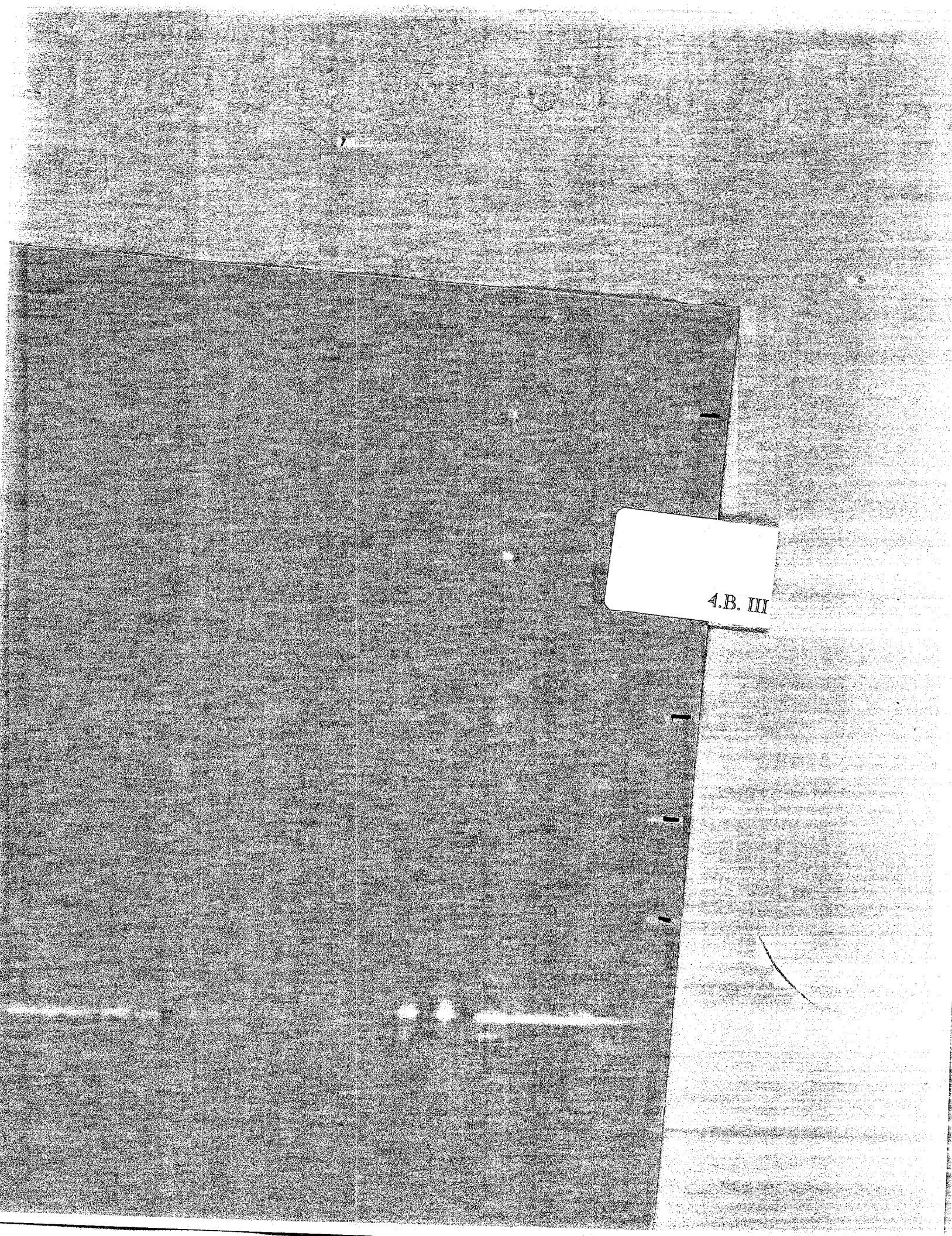


PHOTO 14 **Adrenal cortex:** 3000mg/kg/day×26 weeks, female, ×100, No significant change (Not different from treated group of 3000mg/kg/day×13 weeks and control group)


4.B. III

Safety of Cultured *Agaricus blazei* Murrill (Iwade Strain 101) (Japanese name ; Himematsutake) Preparation, ABME, for Humans in Relatively Long Term Oral Administration.

Shiro Suzuki, M.D., Ph.D.
Tsu Health Clinic
799-7, Kannonji-cho, Tsu, Mie, 514-0062, JAPAN

IWADE RESEARCH INSTITUTE OF MYCOLOGY CO.,LTD.
1-9, SUEHIRO-CHO, TSU, MIE, 514-0012, JAPAN TEL : +81 59 228 5786/FAX : +81 59 224 4661 [e-mail]iwaderim@ztv.ne.jp

November, 17, 1999

Letter of Confirmation

To whom it may concern:

This is to confirm that the test substance, ABME (*Agaricus blazei* Murrill Extract) prepared from cultured *Agaricus blazei* Murrill[Iwade Strain 101], (Japanese name; "Himematsutake"), used for 12-week human study, was identical with the substance, ABME (*Agaricus blazei* Murrill Extract), which has been applied by Iwade Research Institute of Mycology Co., Ltd., as a new dietary ingredient to Food and Drug Administration in the U.S.A.

This is also to confirm that the above mentioned 12-week human study was conducted by Dr. Shiro Suzuki, former professor of 3rd Internal Medicine Department at Mie University School of Medicine, who currently practices at Tsu Health Clinic in Mie, Japan.

Toshimitsu Sumiya

Toshimitsu Sumiya
President
Iwade Research Institute
of Mycology Co., Ltd.
1-9 Suehiro-cho, Tsu
Mie-pref., Japan

Introduction

Several kinds of mushrooms have been used for the maintenance of health or therapy for some disease, for instance, Kofuki Sarunokoshikake or Maitake have been used as diuretics or carcinostatic substance. Among these mushrooms, Japanese researchers, Dr. Iwade et al., noticed that *Agaricus blazei* Murrill (Iwade Strain 101) had most potent anticancer activity in animal experiment and that the nature of the activity was immune modifying one by its polysaccharide, D-glucan. Recently new method to extract D-glucan rich fraction from *Agaricus blazei* Murrill Strain was developed, and resultant extract was named ABME (*Agaricus blazei* Murrill Extract). This report describes the safety of ABME for humans in relatively long term, 12 weeks, oral administration.

Methods of study

ABME is mucous dark brown fluid. Nine persons administered orally daily dose of 30ml of ABME 3 times a day, 10 ml each at morning before breakfast, after lunch, and at night before sleep, for 12 weeks. At the beginning of the test and every other week thereafter, blood pressure estimation, urinalysis, and hematological and biochemical examination of the blood were undertaken. Measurement of body weight was performed at the beginning and the end of the test. Questionnaire for any complaints during administration was done every other week. Nine persons, 4 males and 5 females with age range from 29 to 67, were explained the details of test schedules and the purpose of the test, then all persons gave consent to enroll the test. Table Ⅰ showed the age and sex of nine persons including body weight and other special feature if any. Table Ⅱ showed the testing items. All the items were tested at the beginning and every 2 weeks thereafter, but HbA1c estimation was performed in 4 weeks interval.

Results

Subjective complaints and general condition

During the course of study any complaints atributing ABME administration were not observed. Body weight and blood pressure estimation and urinalysis showed no significant changes during the observation period.

Results of laboratory investigation

Serum total protein and albumin

The change of serum total protein was showed in Fig. 1. All the data were within normal range and no specific change was observed during administration. Serum albumin levels (Fig. 2) were also the same showing no special change during

observation period.

The change of ZTT levels was shown in Fig. 3. There are no special changes during administration indicating no special change of serum protein subfraction.

The change of serum enzyme levels such as GOT, GPT, LDH, ALP, LAP, γ-GTP were shown in Fig. 4, 5, 6, 7, 8, 9. All the data showed no adverse effect of ABME on liver function. Rather GPT and γ-GTP of 67-year-old male person having diabetes showed improving during ABME administration.

The change of serum lipids such as total cholesterol and triglyceride showed in Fig. 10, 11, and there was no special change during administration, i.e. not only persons having initial hyperlipidemia but also persons having normal range of lipids showed no special change during observation period.

The change of BUN, creatinine and uric acid were shown in Fig. 12, 13, 14, and there was no significant change of each value during administration. In a case of 34-year-old male slight rise BUN was observed at 10 week's bled, but 2 weeks later it returned to normal suggesting temporary rise due to unknown cause.

The change of HbA1c levels which was estimated every 4 weeks interval was shown in Fig. 15, and also shown no significant change at all including one case of diabetes having elevated HbA1c levels of 6 %.

Hematological data were shown in Fig. 16, 17, 18, 19, 20, each the change of RBC, WBC, hemoglobin, hematocrit, and platelet, indicating no significant change during administration. Leucocyte differential count was also estimated, and no significant change was observed (data not shown).

Conclusion

Oral ABME administration of 30 ml a day for 12 weeks in 9 persons showed no adverse effect on hematology, enzyme biochemistry, urinalysis, and kidney function. Rather initial elevation of GPT and γ-GTP of one case showed improvement during administration. Therefore the safety of oral administration of ABME was confirmed in relatively long term.

Table I Nine persons enrolled to the test

	Sex	Age	Body weight		Blood pressure		Others
			Before	After	Before	After	
K.M.	M	49	65.8	65.7	150/ 91	136/ 89	Low grade hypertension
K.S.	M	67	58.3	58.9	156/ 95	177/100	HT and IGT
H.N.	M	49	82.3	81.0	157/101	147/97	HT and Hyperlipidemia
Y.I.	M	34	81.0	80.9	142/ 90	135/ 81	
A.M.	F	33	60.4	57.9	131/ 78	104/ 47	
K.S.	F	65	46.1	44.5	135/ 83	111/ 68	
S.M.	F	42	54.9	54.0	117/73	102/ 61	
E.S.	F	29	45.8	46.6	93/ 59	106/ 58	
S.K.	F	65	47.5	47.6	132/73	124/ 76	

Table II Tests performed during ABME administration

Blood pressure, Body weight, Urinalysis, Complete blood count and WBC differential count, Total protein, Albumin, A/G, ZTT, ALP, LAP, GOT, GPT, LDH, γ-GTP, Total cholesterol, Triglyceride, BUN, Creatinine, Uric, acid, HbA1c (every 4 weeks)

End of report

Fig. 1 Serum total protein

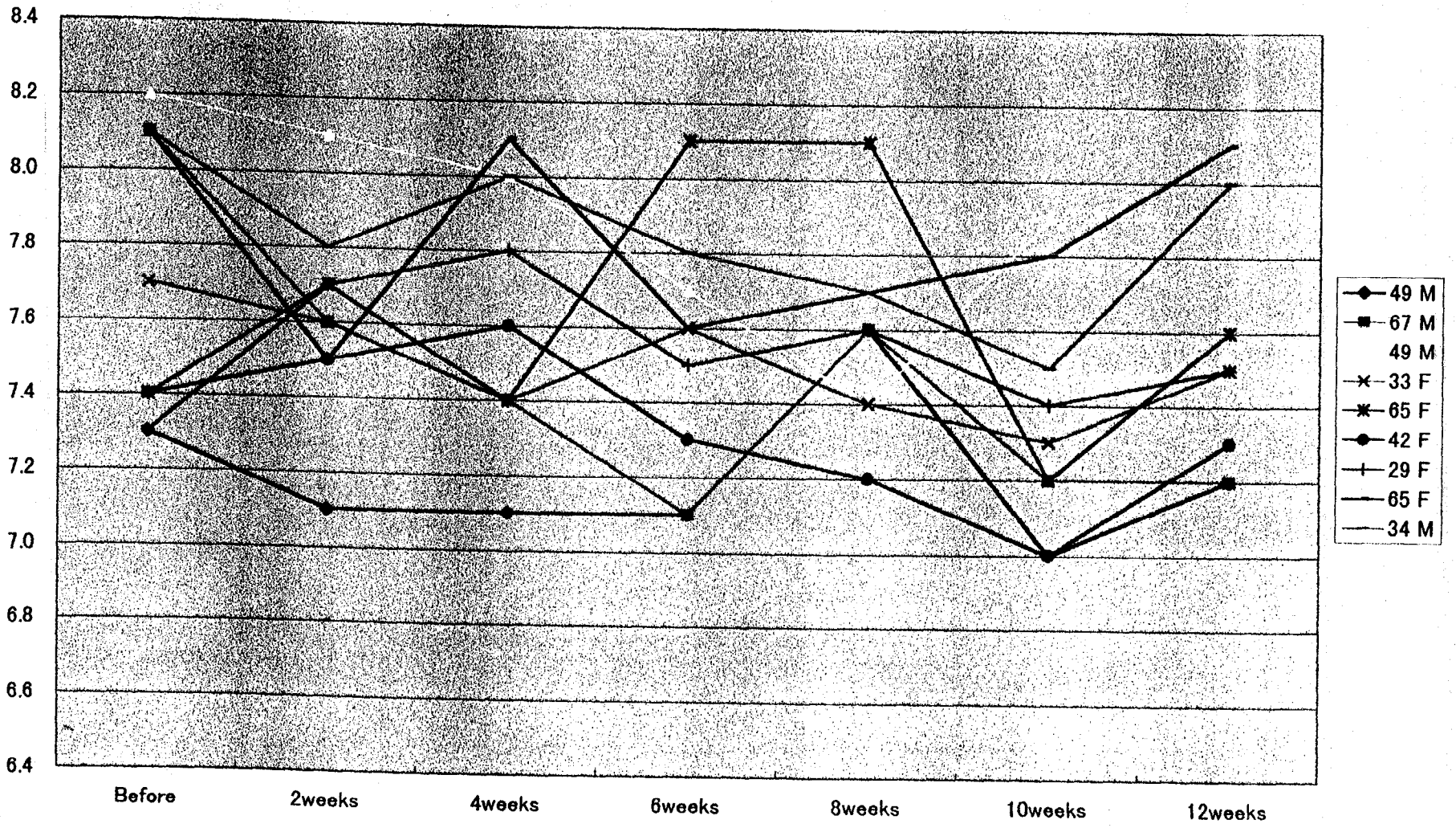


Fig. 2 Albumin

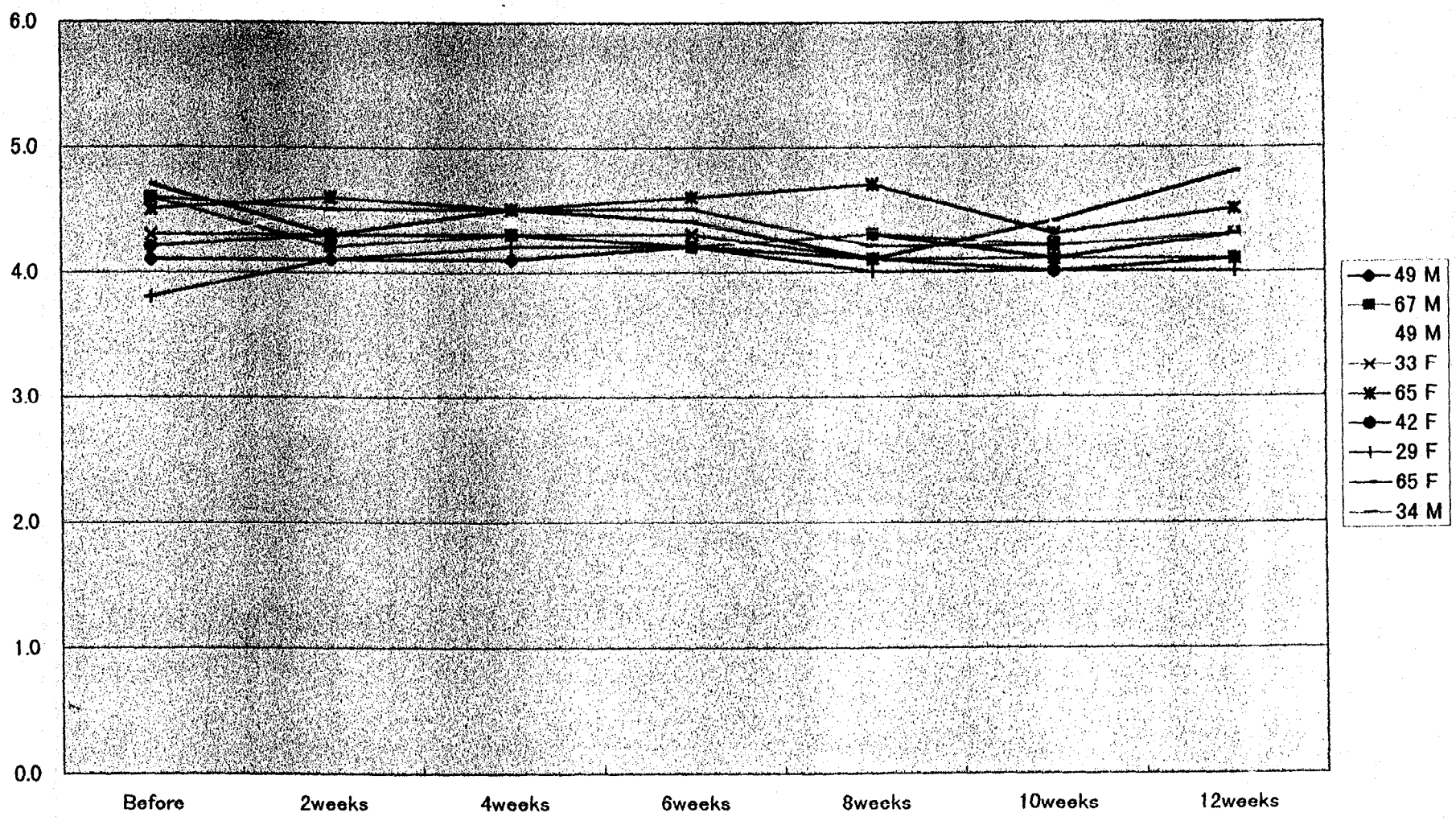


Fig. 3 ZTT

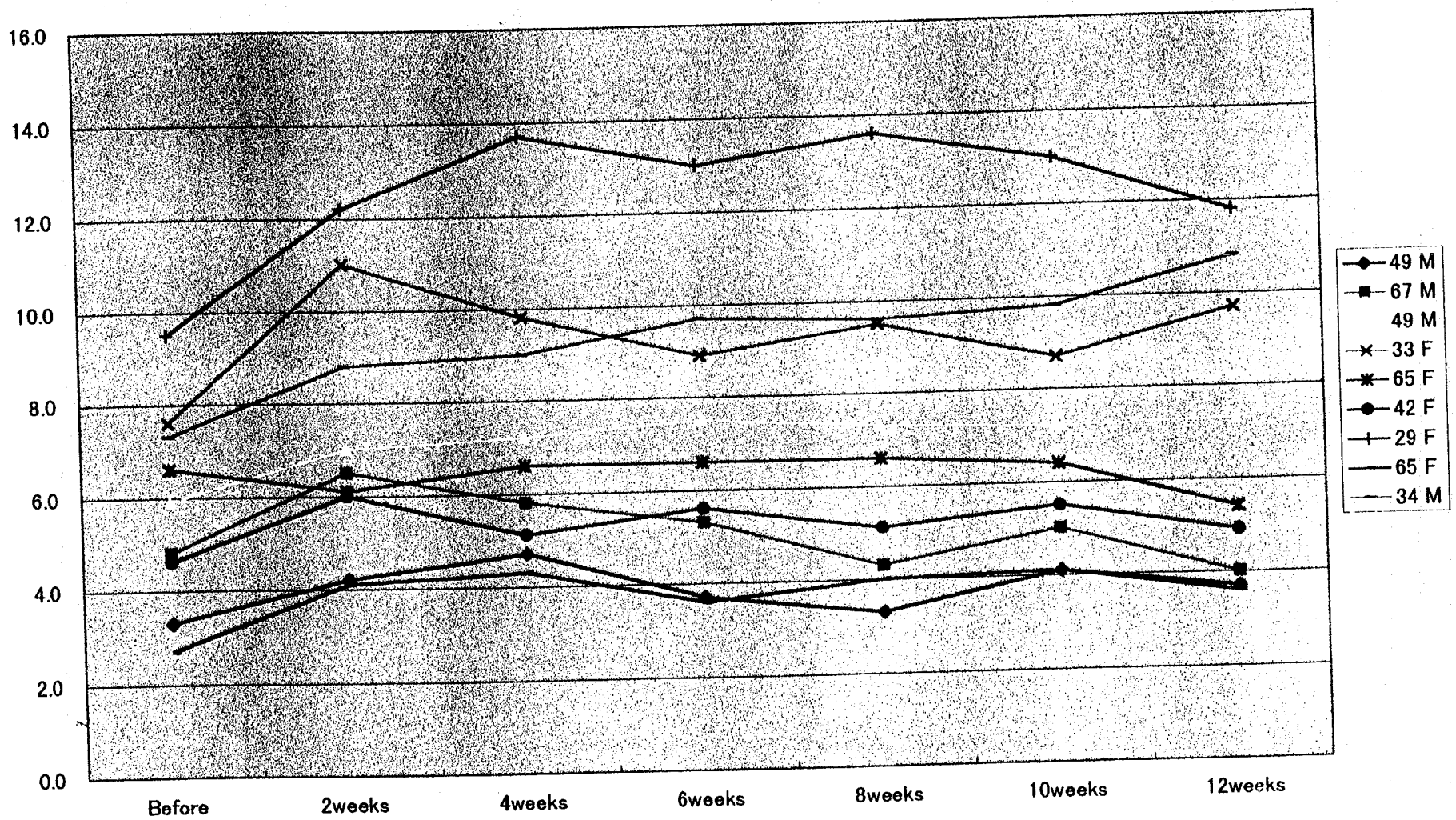

16.0
14.0
12.0
10.0
8.0
6.0
4.0
2.0
0.0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 4 ALP

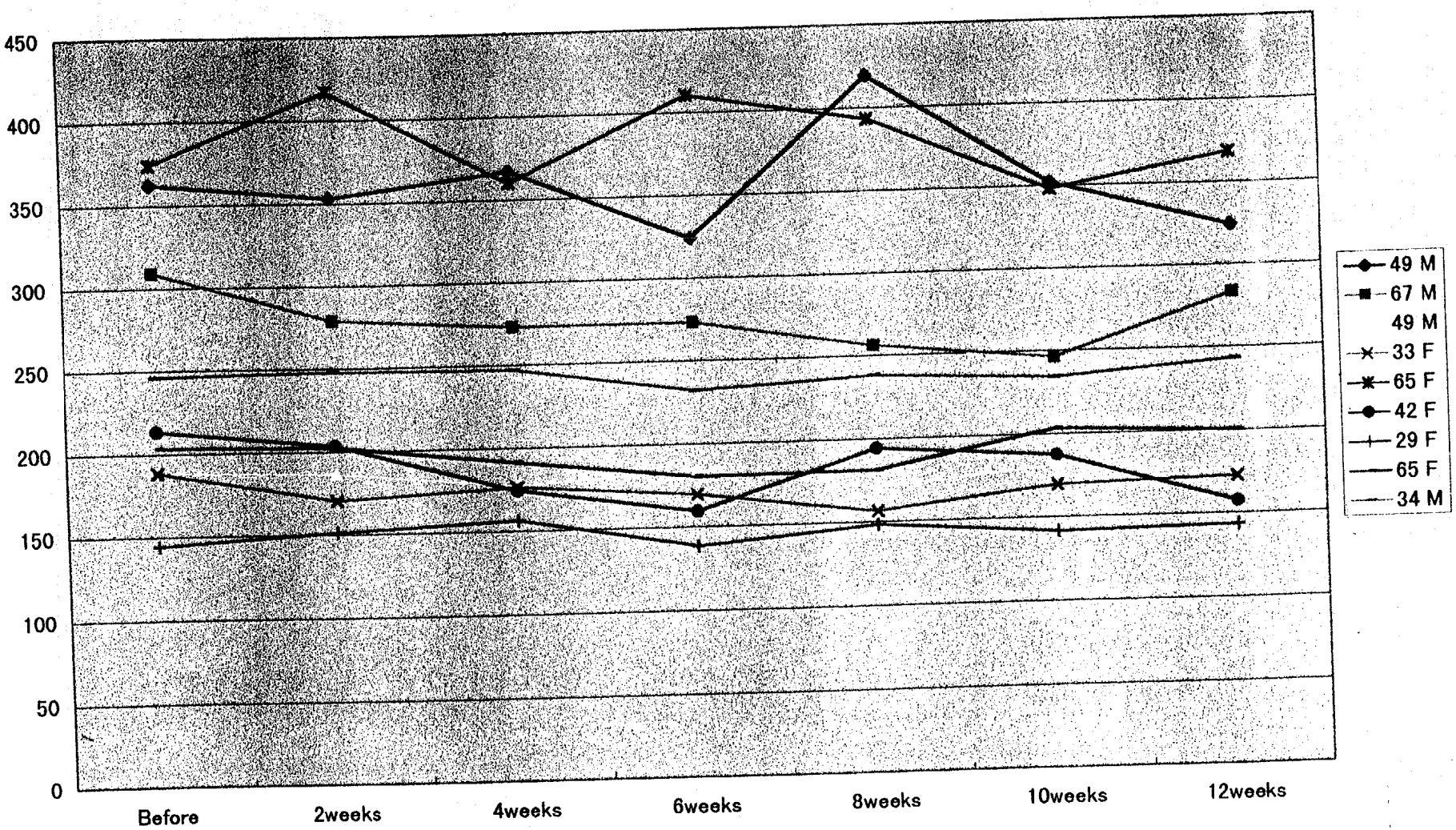

450
400
350
300
250
200
150
100
50
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 5 LAP

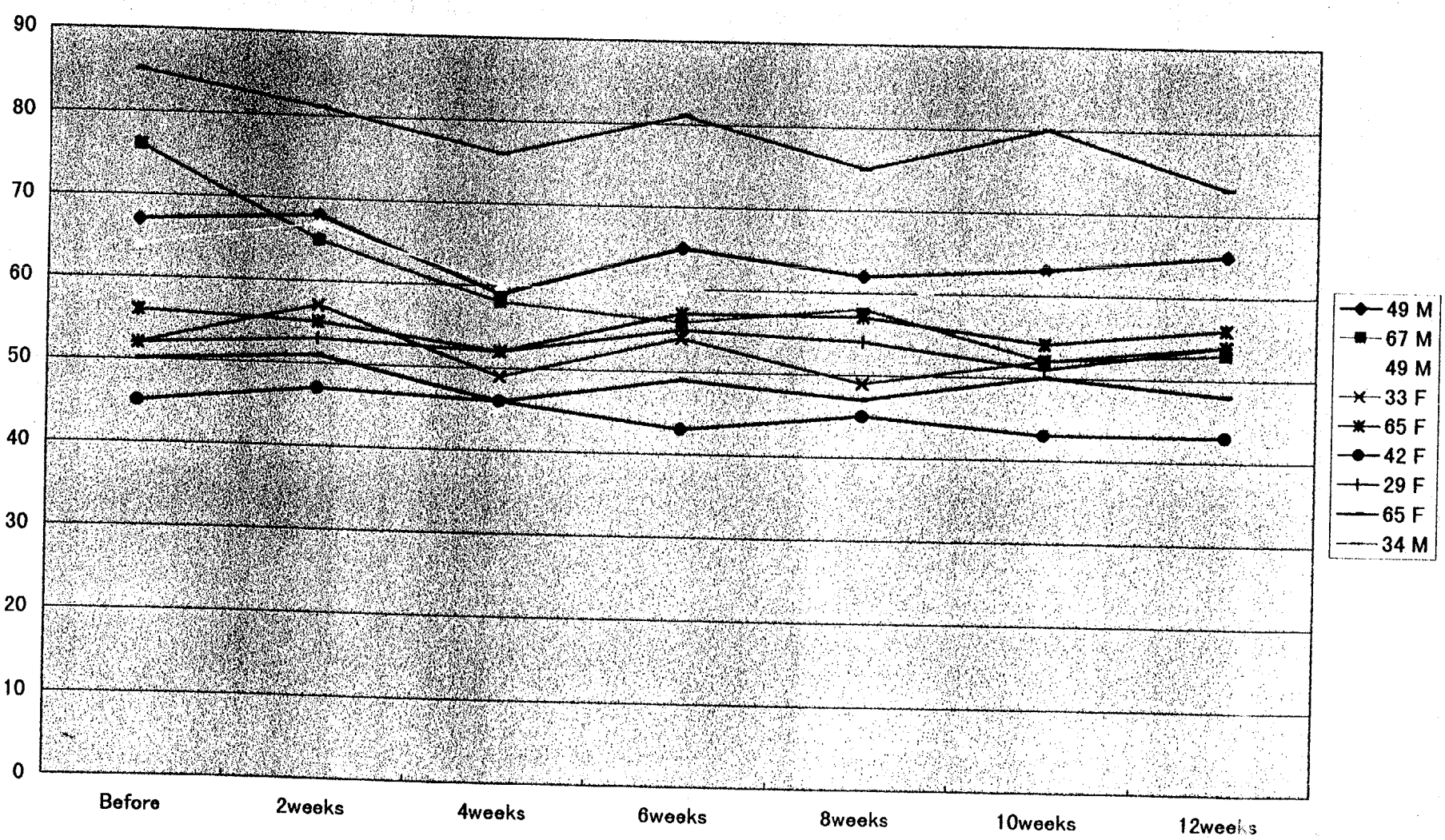

90
80
70
60
50
40
30
20
10
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 6 GOT

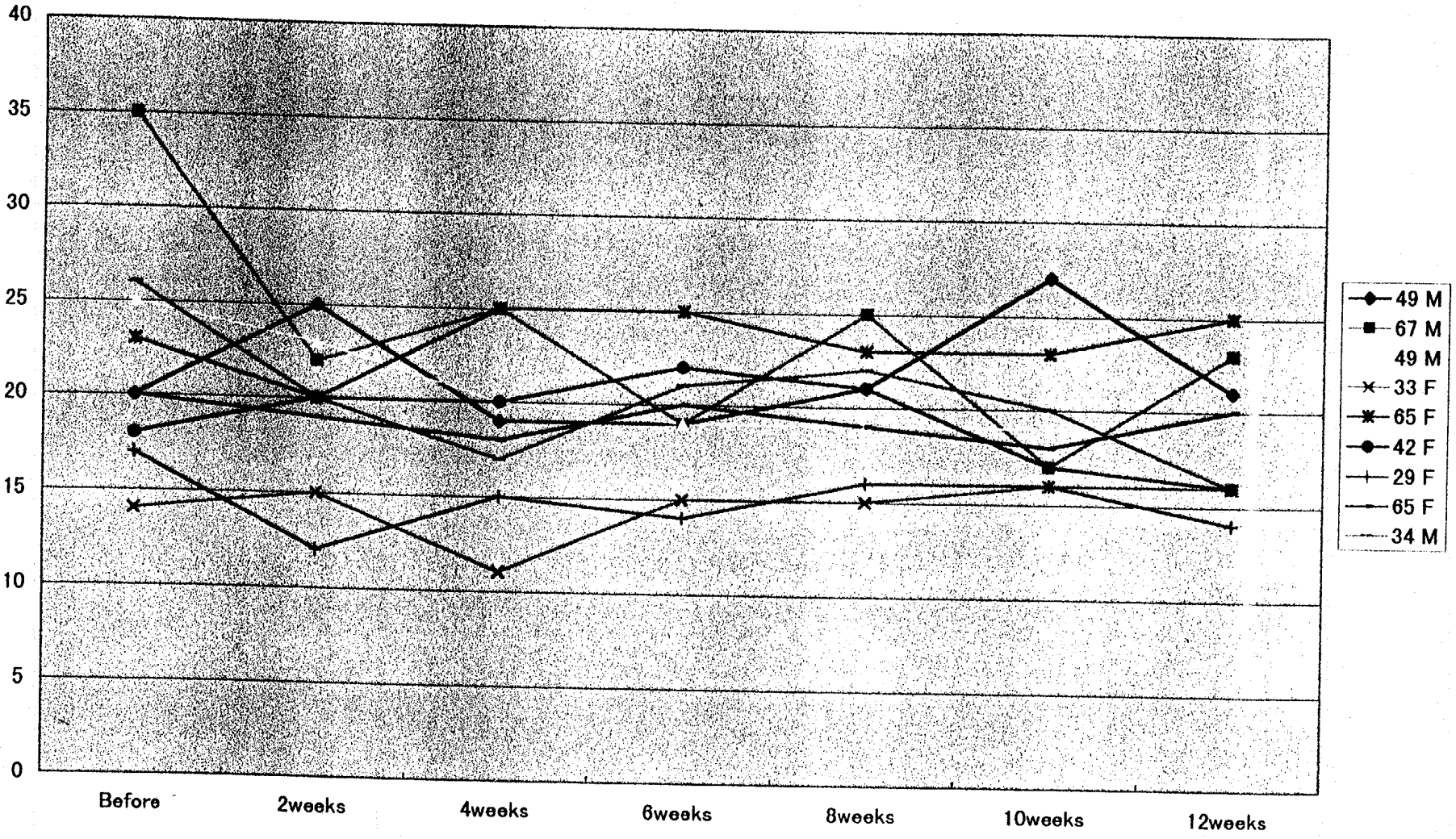

40
35
30
25
20
15
10
5
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 7 GPT

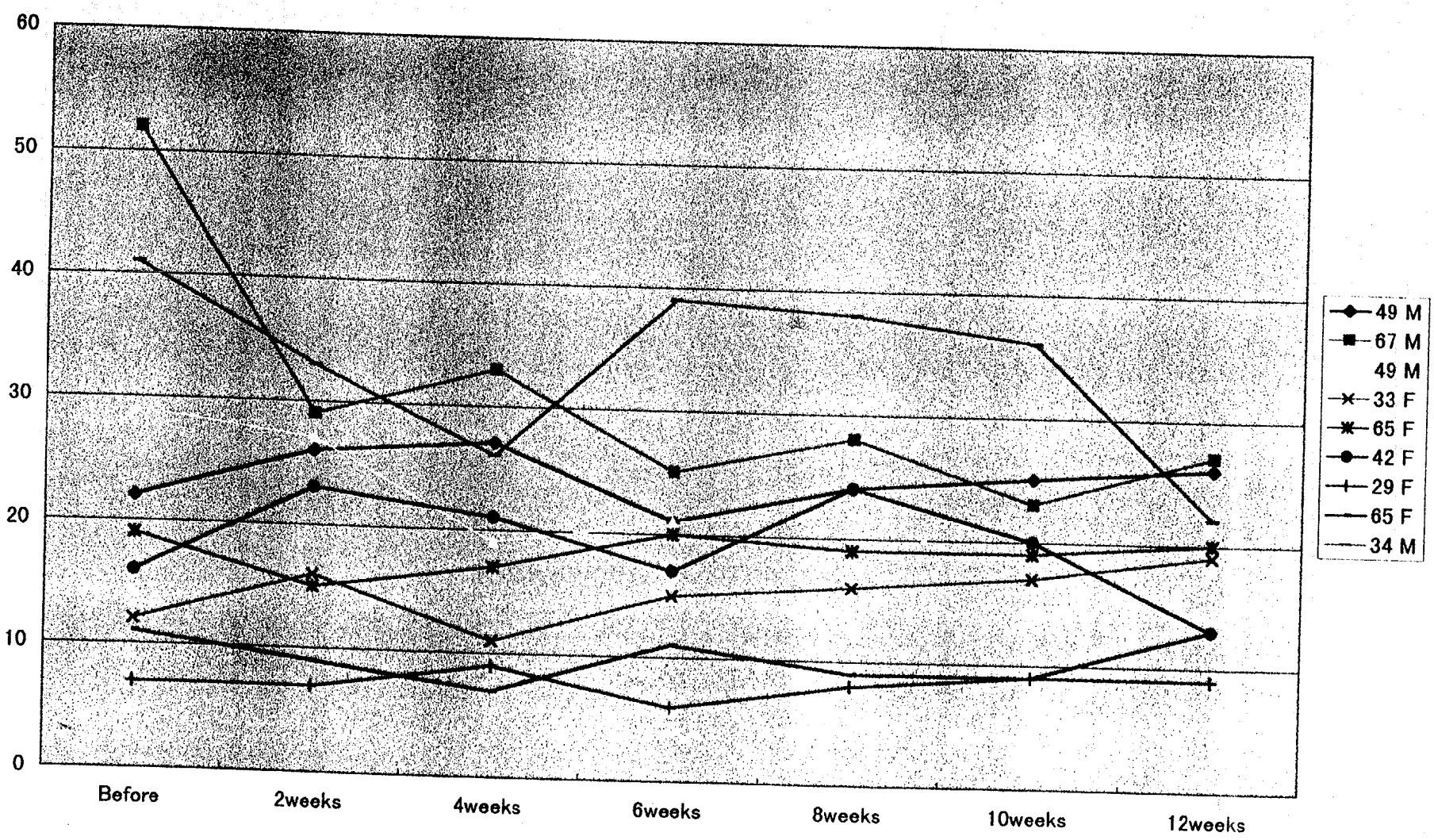

60
50
40
30
20
10
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 8 LDH

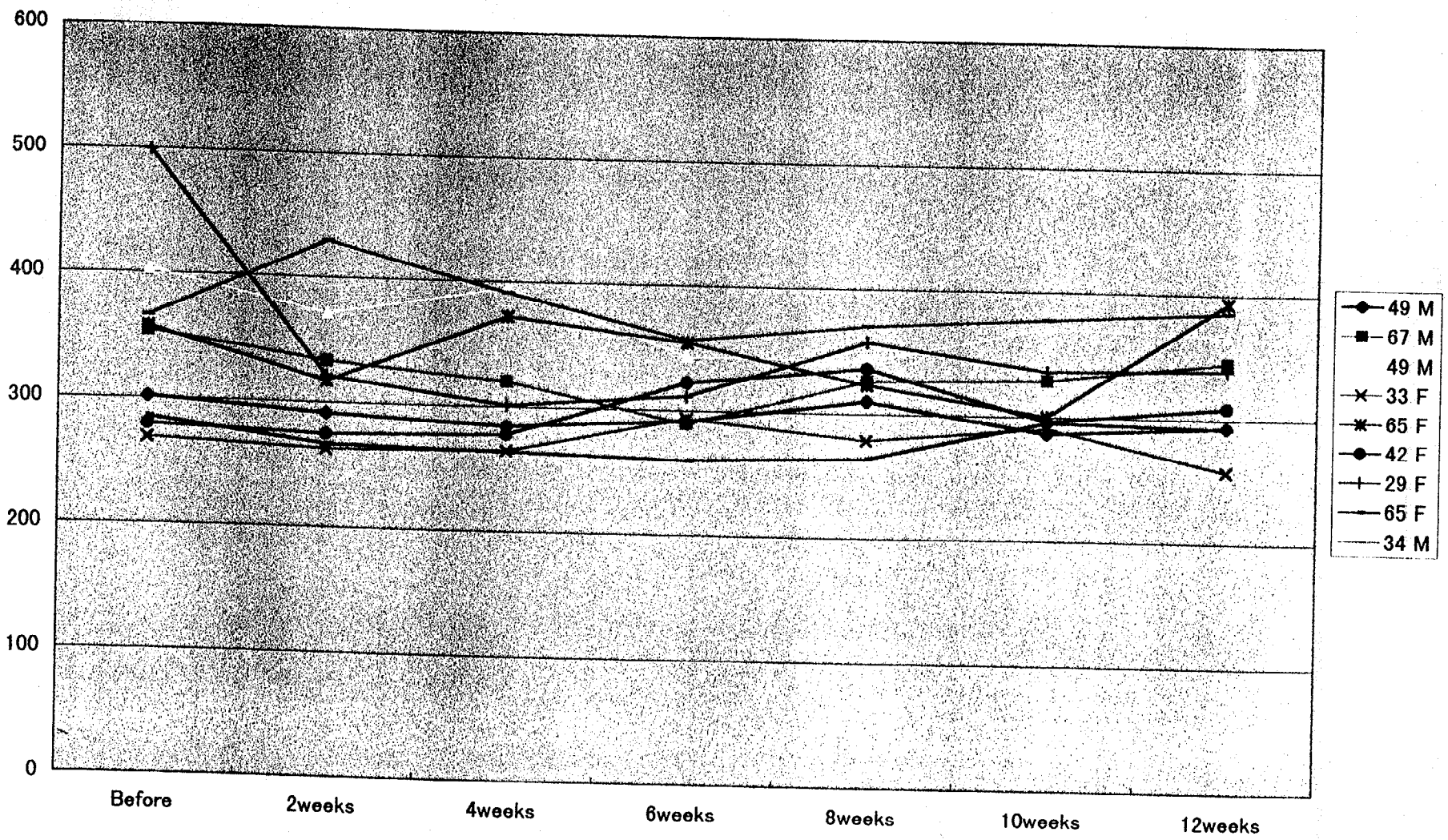

600
500
400
300
200
100
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 9 γ—GTP

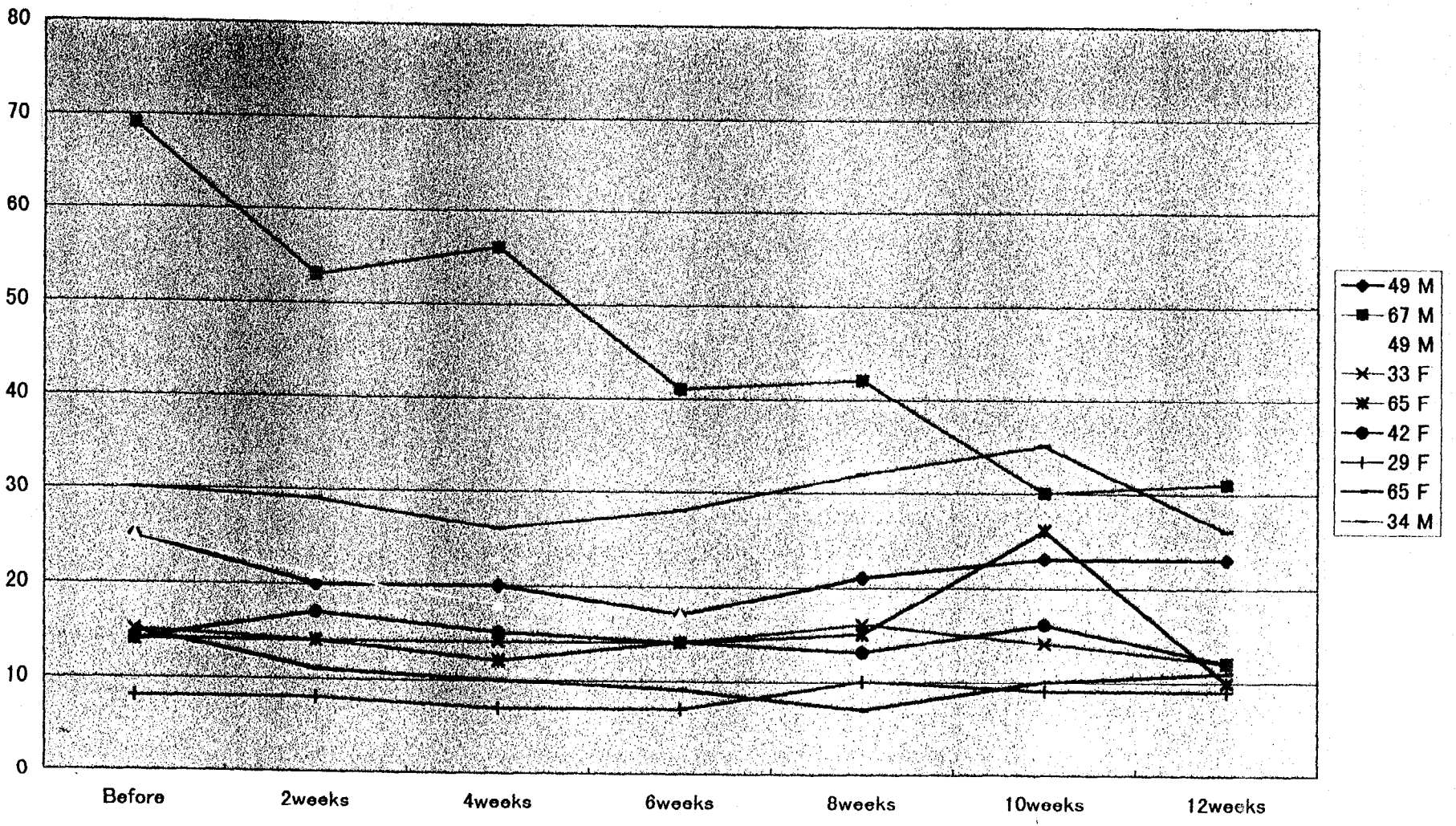

80
70
60
50
40
30
20
10
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 10 Total cholesterol

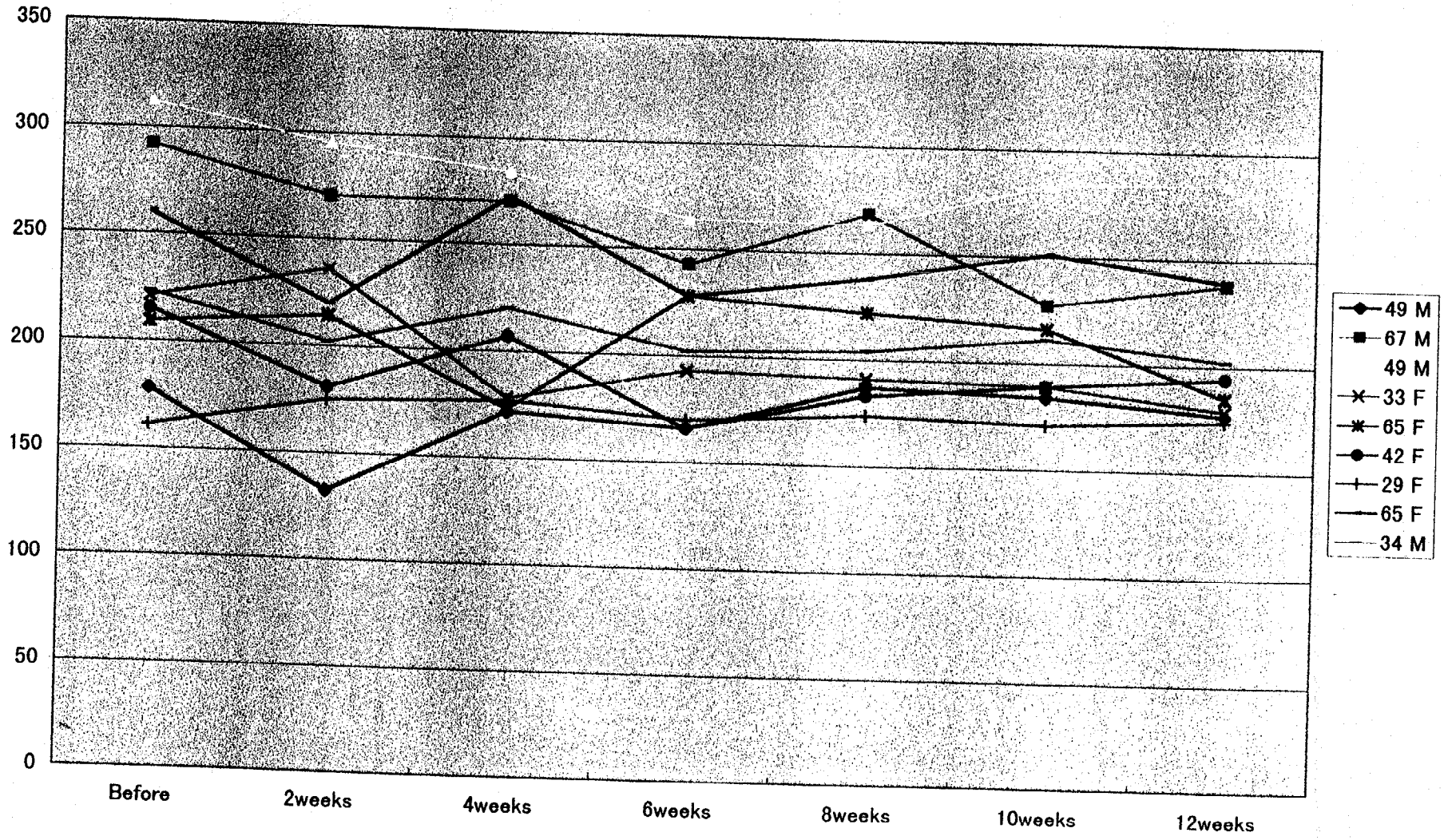


Fig. 11 Triglyceride

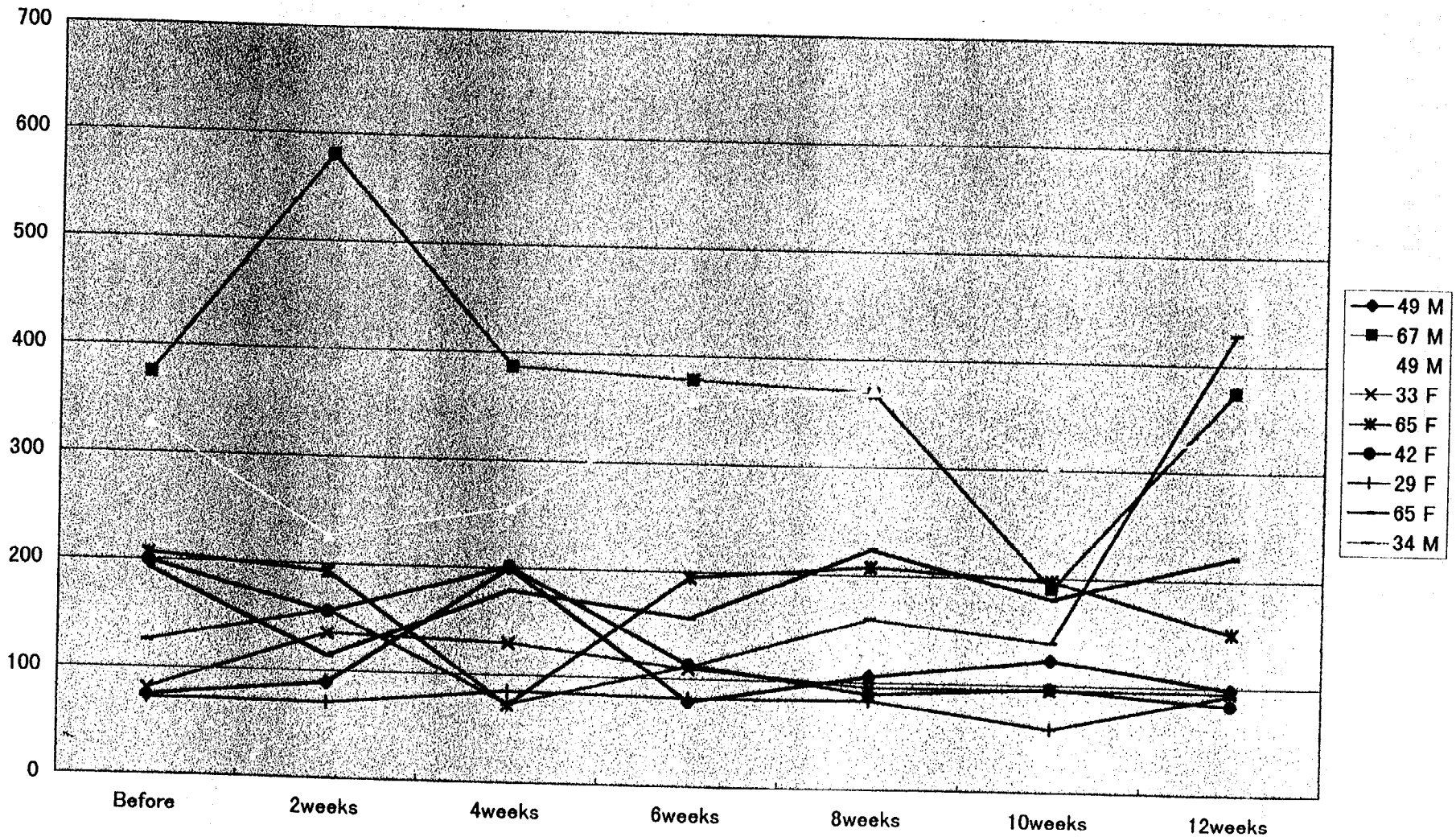


Fig. 12 BUN

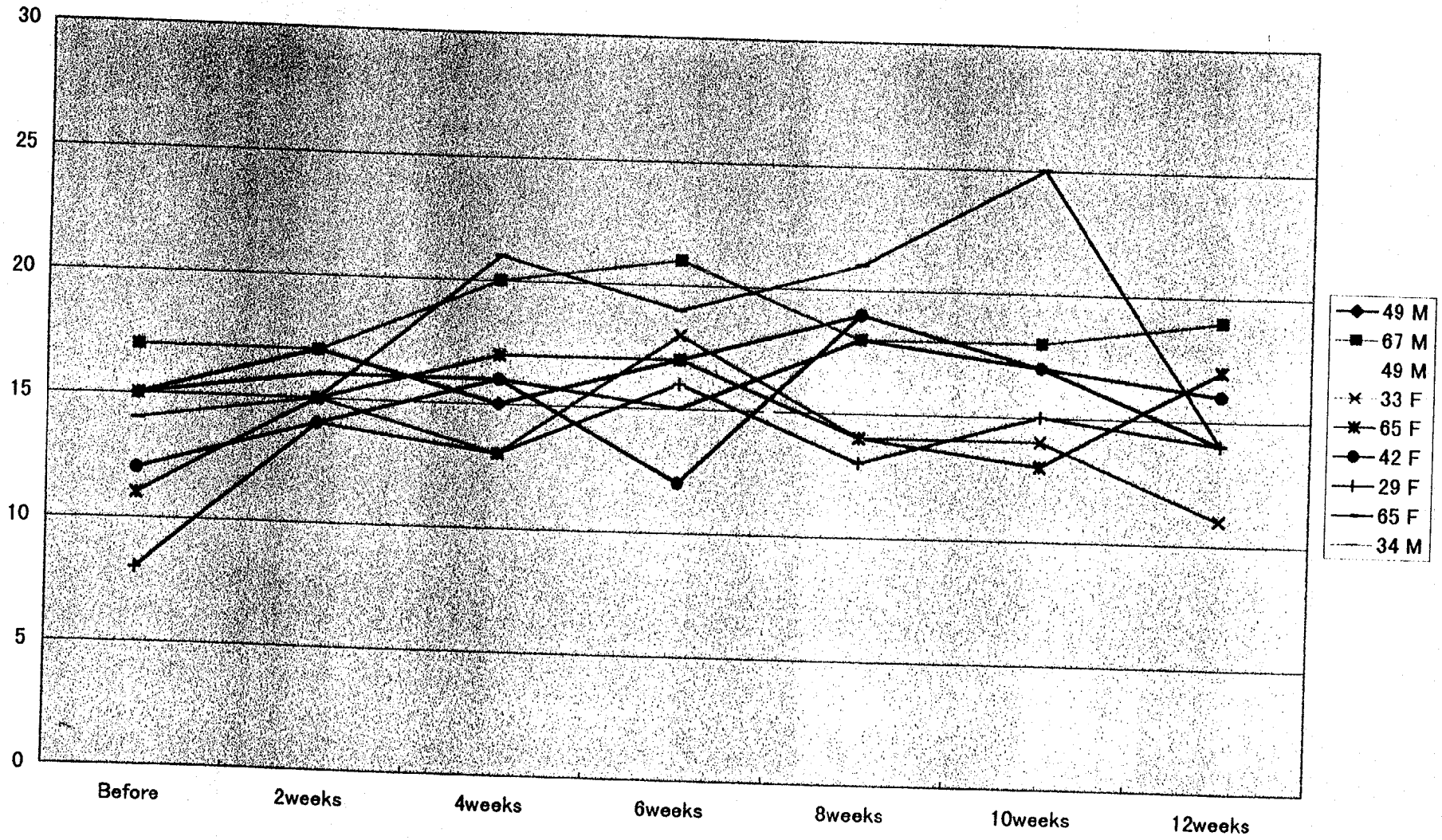

30
25
20
15
10
5
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 13 Creatinine

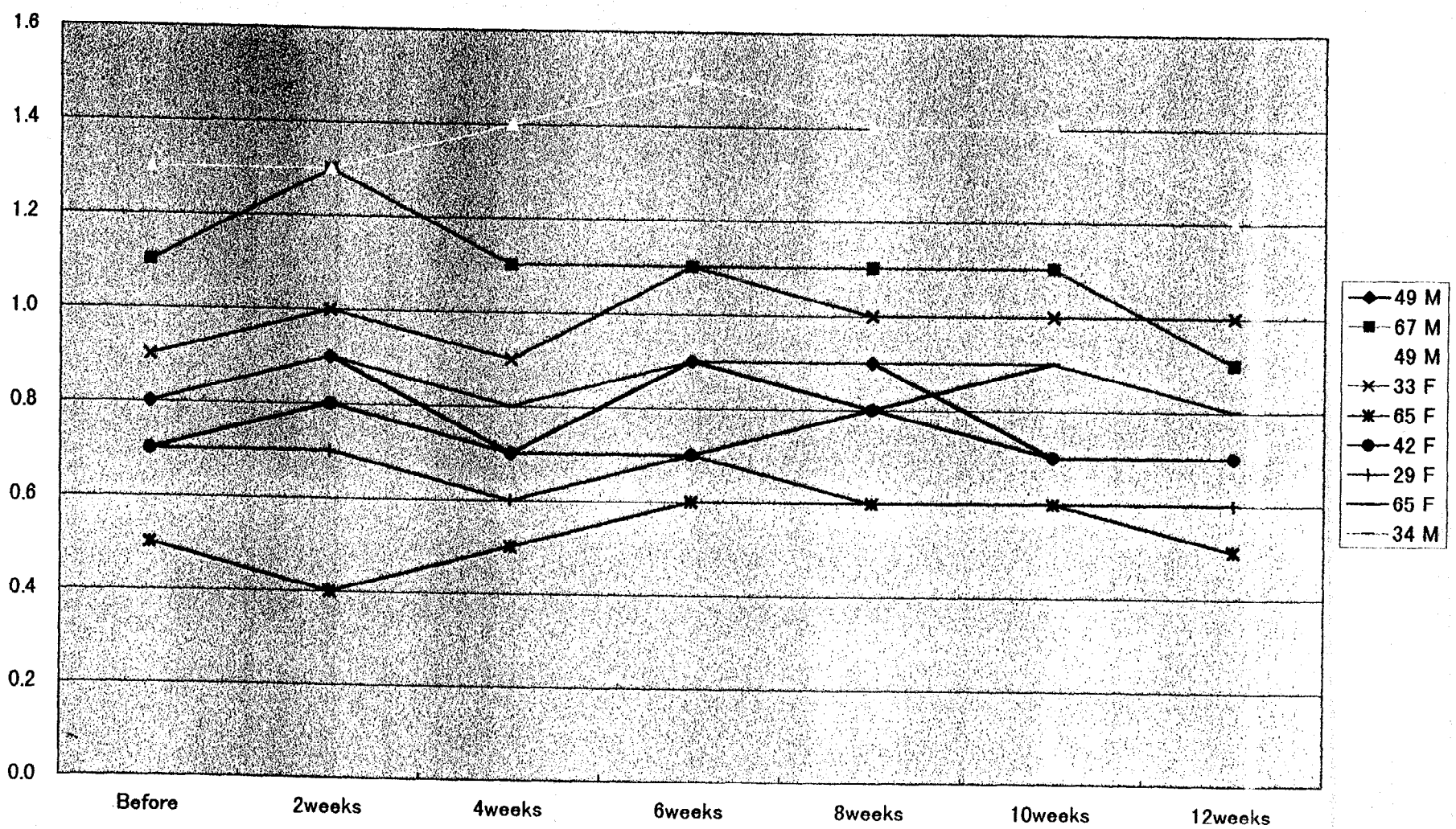


Fig. 14 Uric acid

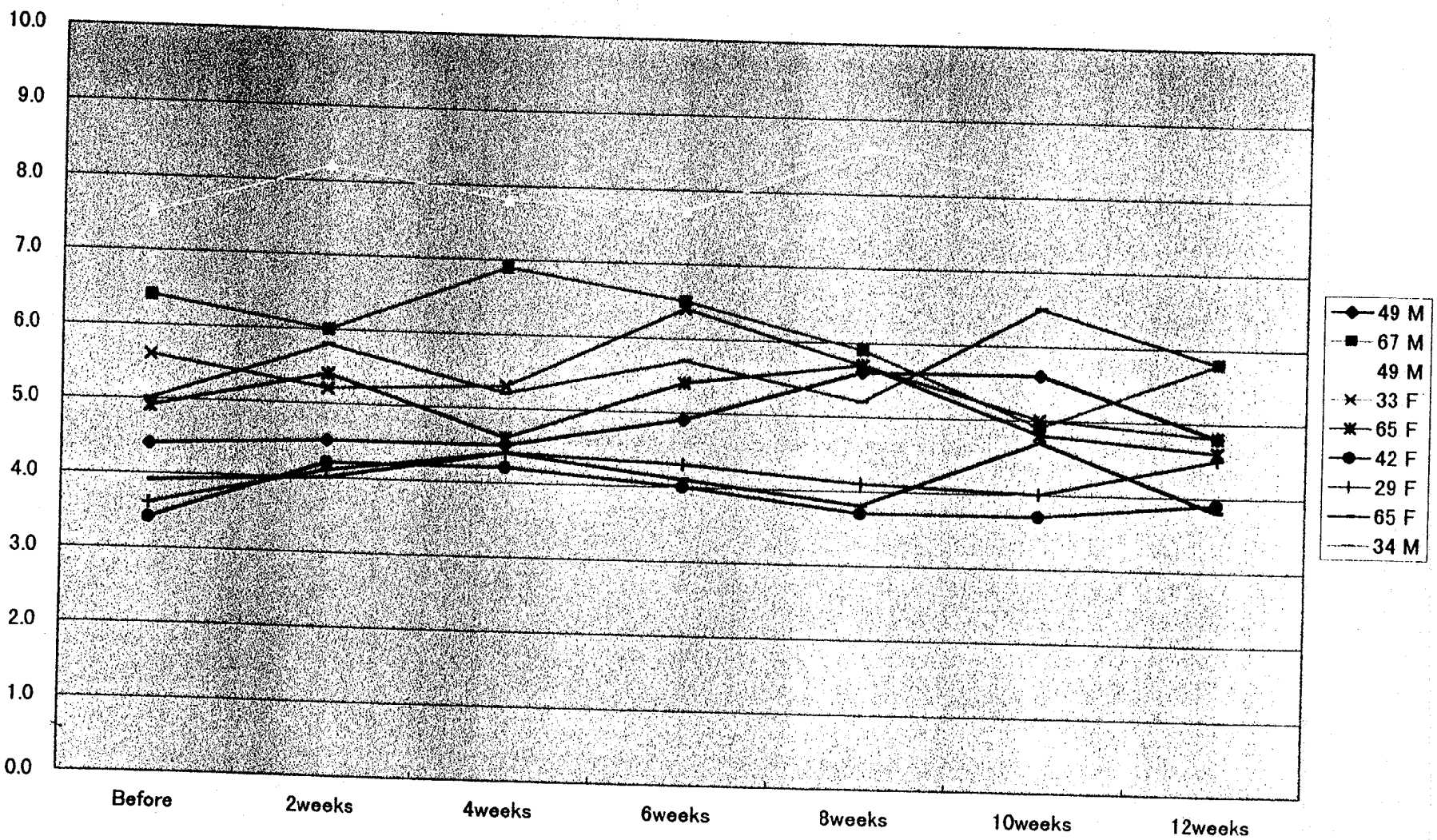


Fig. 15 HbA1c

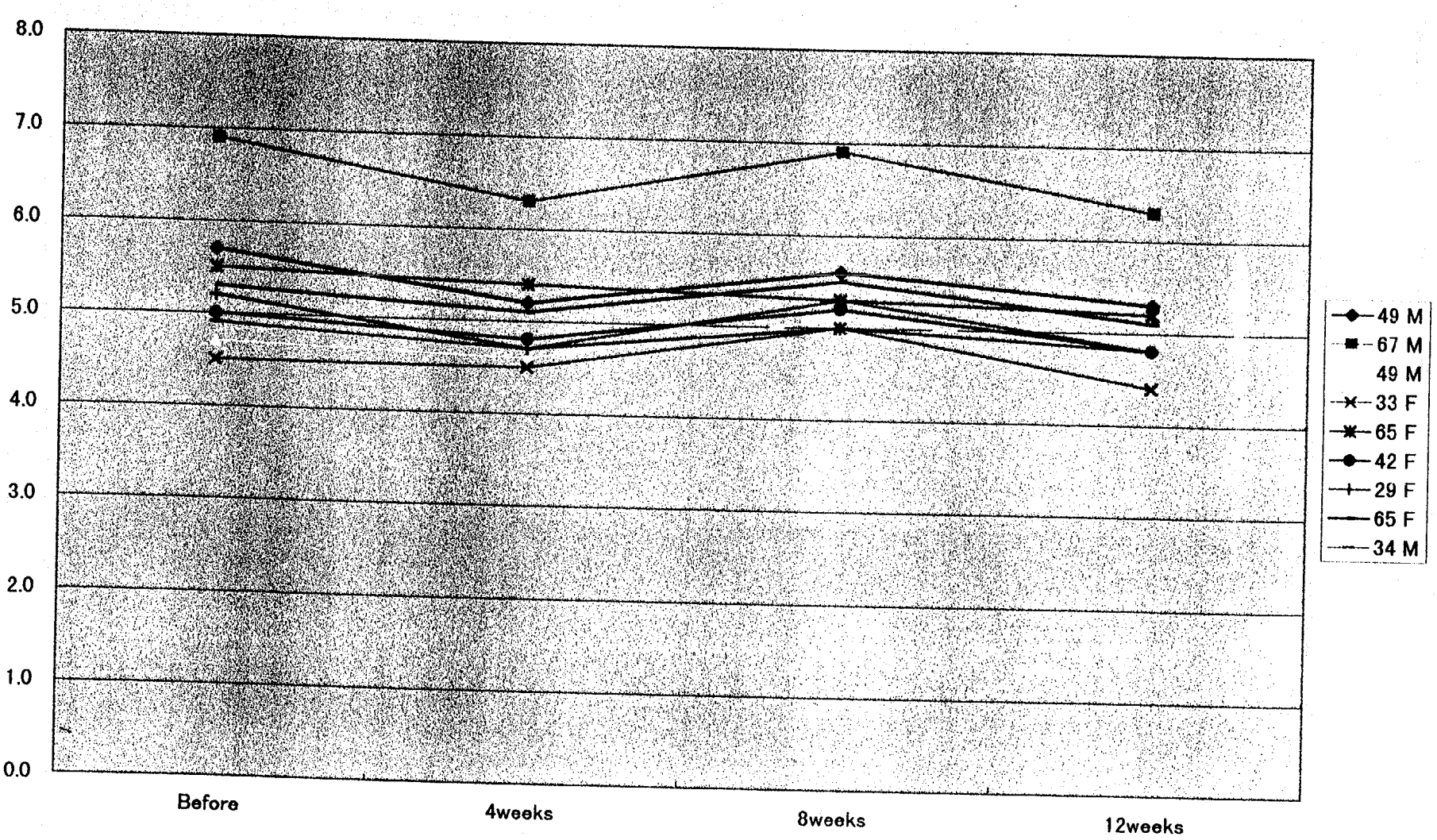

8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
Before
4weeks
8weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 16 WBC

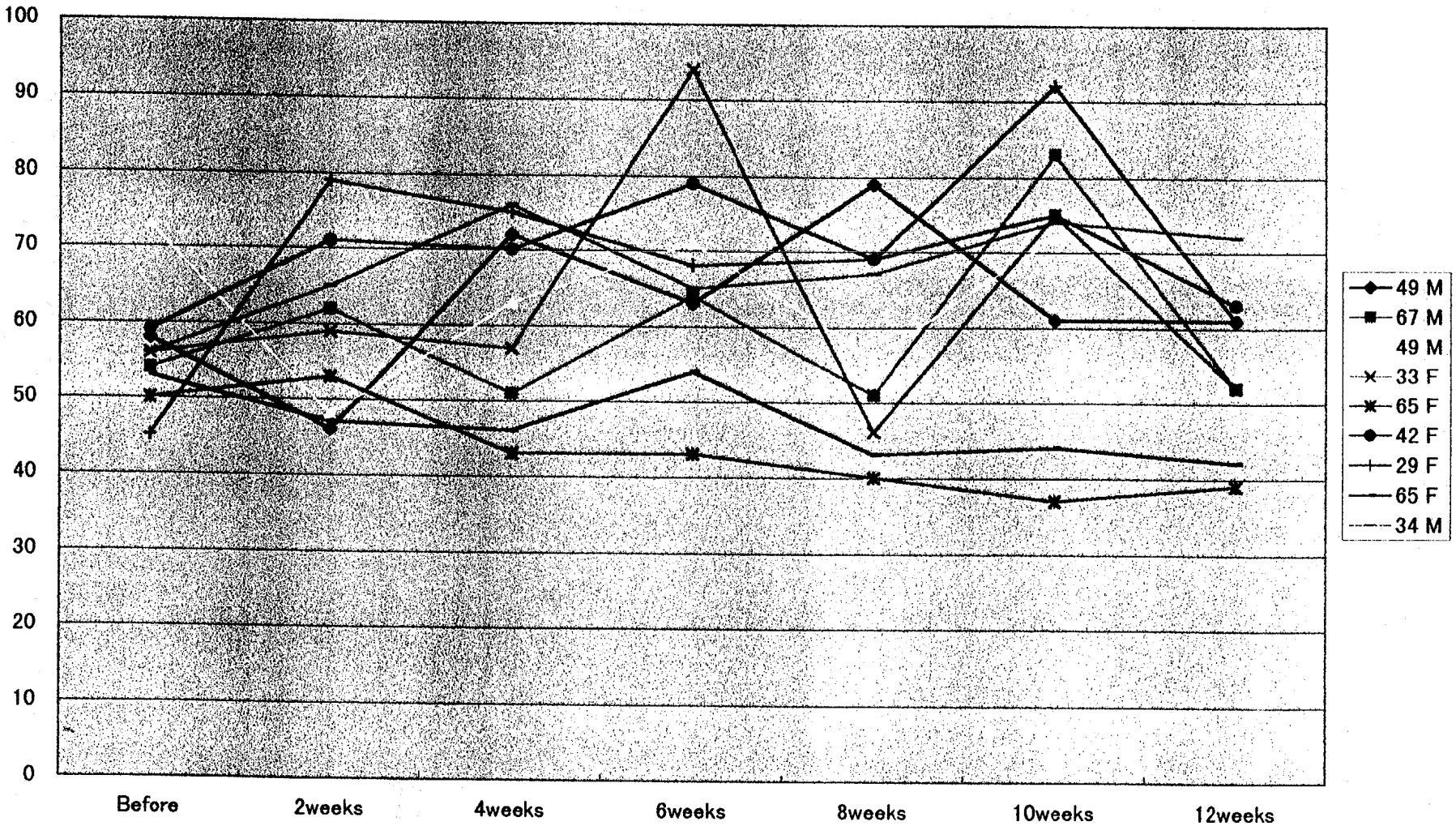

100
90
80
70
60
50
40
30
20
10
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 17 RBC

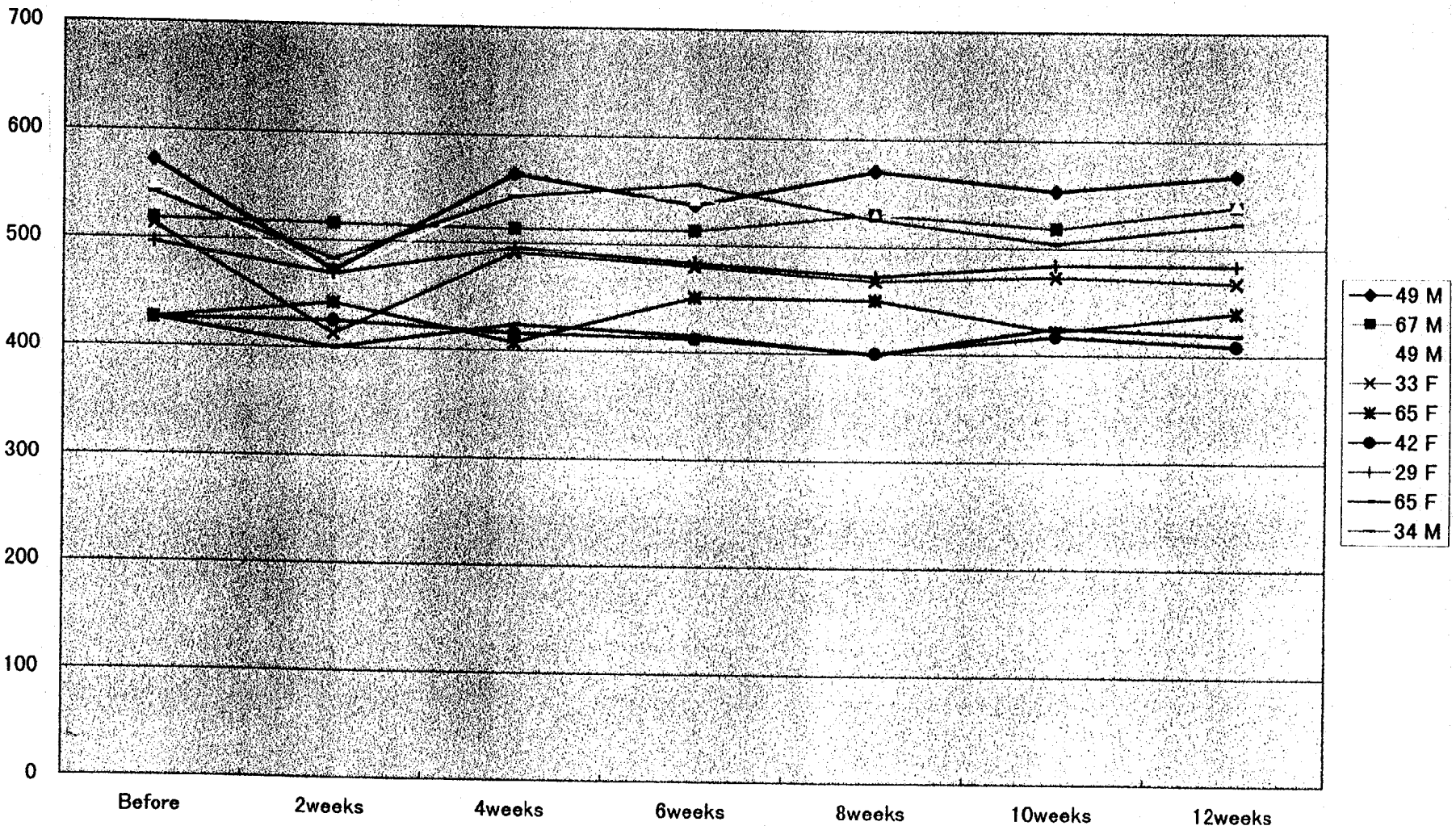

700
600
500
400
300
200
100
0
Before
2weeks
4weeks
6weeks
8weeks
10weeks
12weeks
49 M
67 M
49 M
33 F
65 F
42 F
29 F
65 F
34 M

Fig. 18 Hemoglobin

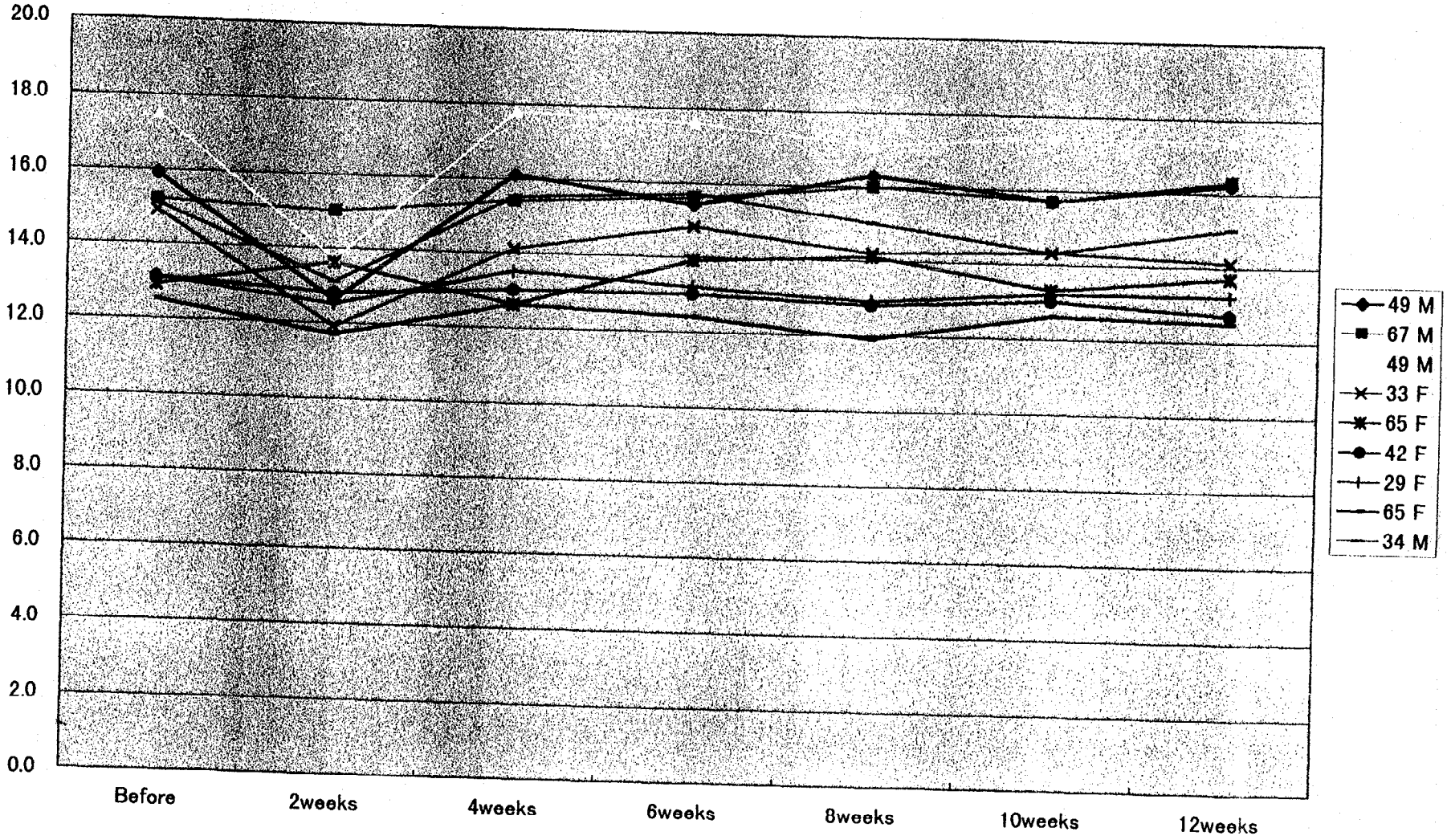


Fig. 19 Hematocrit

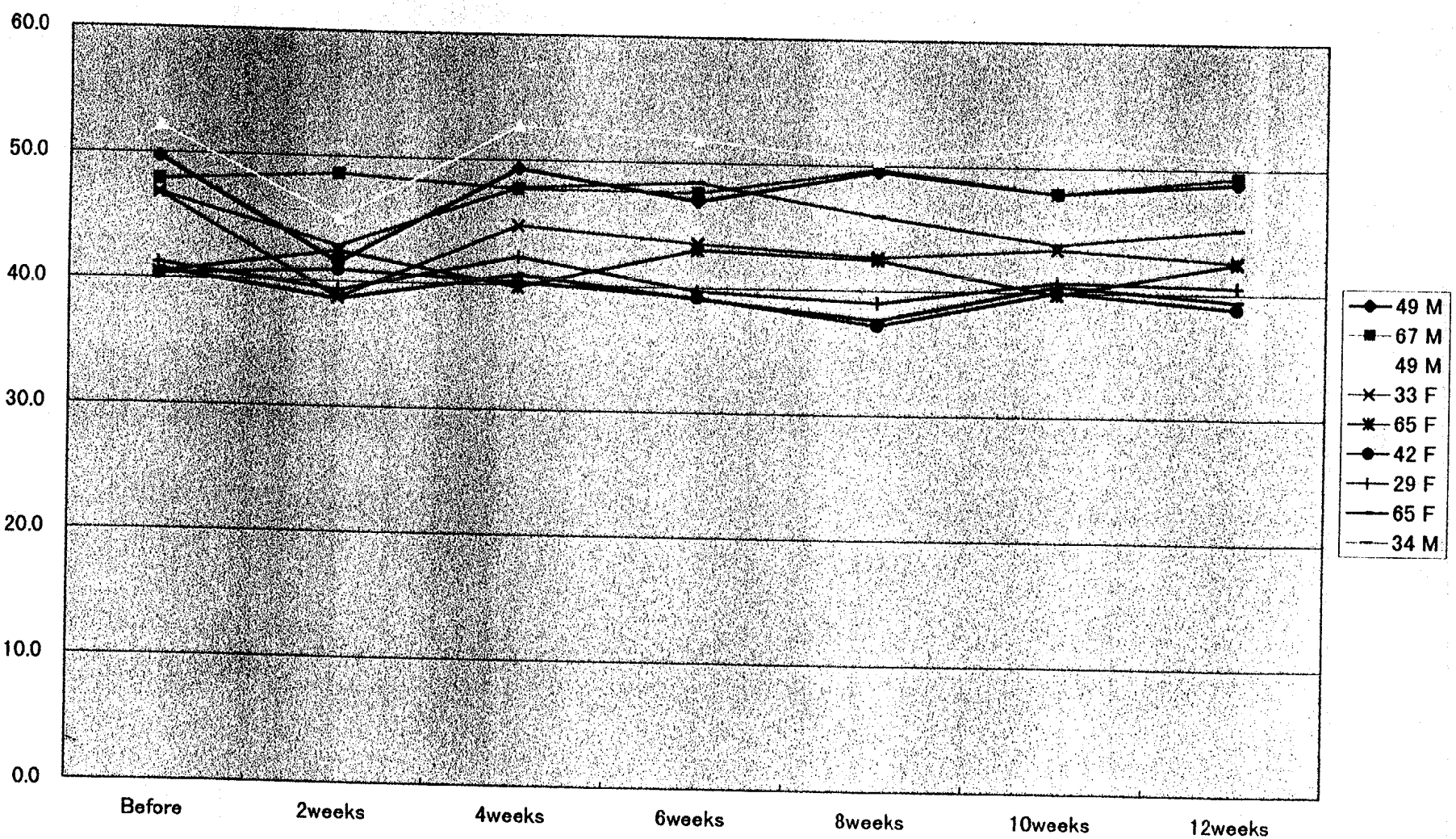
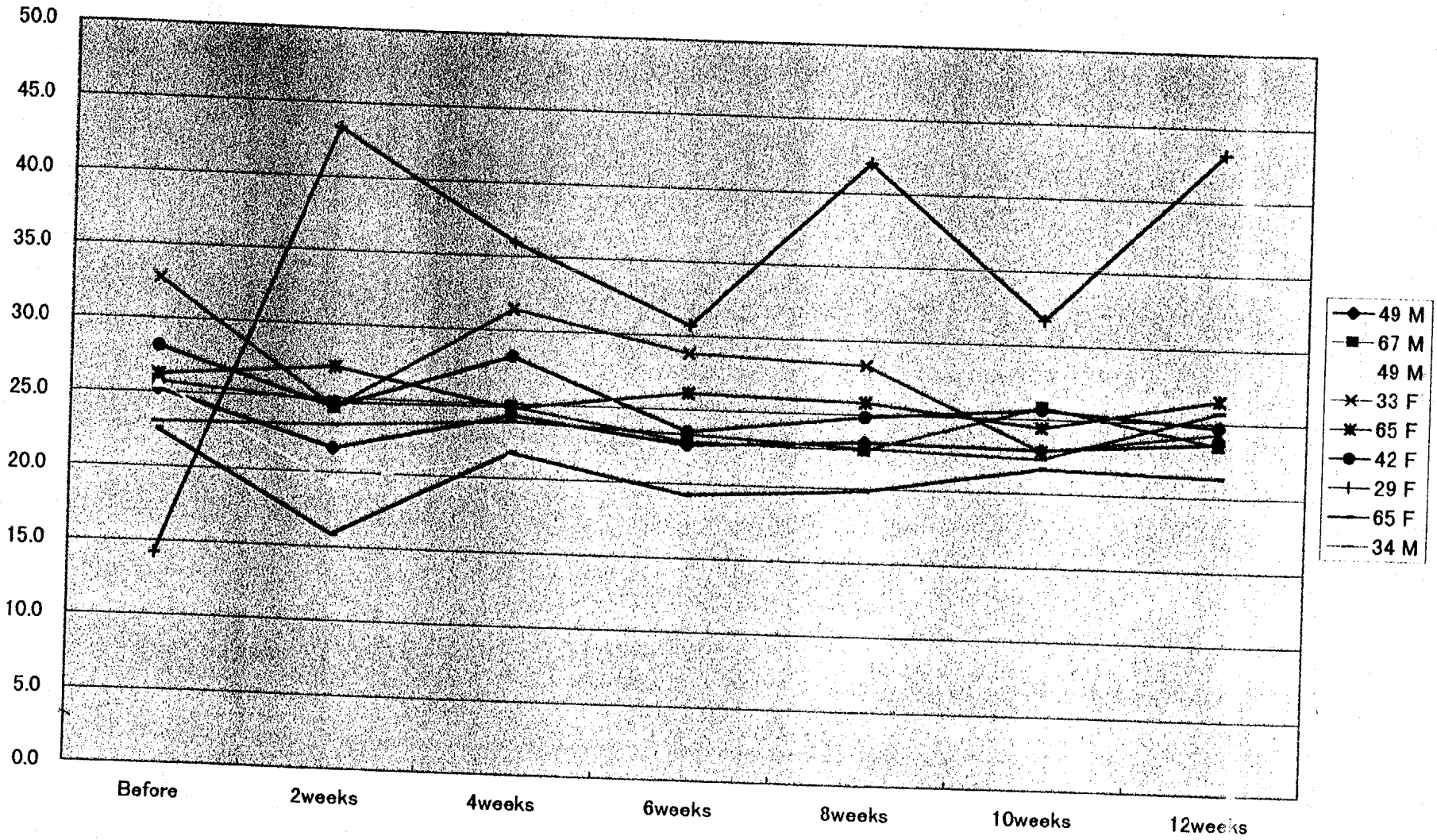


Fig. 20 Platelet



CONFIDENTIAL

[redacted]

検査成績報告書

3389(2377)　津健康クリニック　津健康クリニック　津健康クリニック

IDNO 277 087

[redacted]

[redacted]

性 男性
49才
Male 49
K.M

混濁（1＋）

検査項目	11/03/02 測定値	11/03/16 測定値	11/03/30 測定値	単位	基準値
★血清総蛋白	7.3	7.1	7.1	g/dl	6.5～8.2
★A/G比	1.3	1.4	1.4		1.1～2.0
★アルブミン	4.1	4.1	4.1	g/dl	3.7～5.3
★ZTT	3.3	4.2	4.7	units	3.0～12.0
★ALP	↗ 362	↗ 353	↗ 367	IU/l	100～350
★LAP	67	68	59	IU/l	35～75
★GOT	20	25	19	IU/l	10～40
★GPT	22	26	27	IU/l	6～40
★LDH	301	291	284	IU/l	230～460
★γ-GTP	25	20	20	IU/l	50以下
★総コレステロール	178	132	172	mg/dl	130～220
★トリグリセライド	74	脂 89	↗ 200	mg/dl	35～150
★尿素窒素	15	17	15	mg/dl	8～21
★クレアチニン	0.8	0.9	0.7	mg/dl	0.6～1.3
★尿酸	4.4	4.5	4.5	mg/dl	2.5～7.5
★Hb-A1C	5.7		5.2	%	4.3～5.8
		46			
★白血球数	58	472	72	×10³/μl	36～92
★赤血球数	↗ 572	↘ 12.6	↗ 564	×10⁴/μl	420～560
★ヘモグロビン量	15.9	41.5	16.1	g/dl	13.0～17.0
★ヘマトクリット値	49.7	88	49.3	%	39.0～50.0
MCV	87	↘ 26.7	87	fl	81～98
MCH	27.8	↓ 30.4	28.5	pg	27.0～33.5
MCHC	32.0	21.6	32.7	%	32.0～35.0
★血小板数	25.2		24.2	×10⁴/μl	14.0～35.0
		0.0			
		0.0	0.0	%	0.0
		42.5	0.0	%	0.0
		3.9	52.3	%	36.0～69.0
		1.1	4.1	%	1.0～5.0
		45.8	0.7	%	0.0～2.0
		0.0	37.9	%	27.0～53.0
		6.7	0.0	%	0.0
			5.0	%	2.0～10.0

総合報告書Ⅰ　受付 11/03/0　受付 11/03/3　受付 11/03/30　報告 11/03/31

登録衛生検査　登録衛生検査所　登録衛生検査所　実施料 329点

担当者 大谷 敦

(株)メディック　(株)メディック　(株)メディック

TEL(059)2　TEL(059)2　TEL(059)224-5455

[redacted]　検査成績報告書

CONFIDENTIAL

検査成績報告書

検査成績報告 検査成績報告 検査成績報告書

津健康クリニック 津健康クリニック 津健康クリニック 様

3389(2377) 津健康クリニック [redacted] 3389(2377)

[redacted] 男性 50才

	実施料 329点	実施料 244点	実施料 329点			
	IDNo 2-77009	IDNo 2-77082	IDNo 2-77092			
検査項目	受付11/04/13 測定値	受付11/04/27 測定値	受付11/05/11 測定値	受付11/05/25 測定値	単位	基準値	報告11/05/26 検査項目
★血清総蛋白	7.1	7.6	7.0	7.2	g/dl	6.5～8.2	*血清総蛋白
★A/G比	1.4	1.3	1.4	1.5		1.1～2.0	*A/G比
★アルブミン	4.2	4.3	4.1	4.3	g/dl	3.7～5.3	*アルブミン
★ZTT	3.7	3.3	4.1	3.7	units	3.0～12.0	*ZTT
★ALP	324	419	352	324	IU/l	100～350	*ALP
★LAP	65	62	63	65	IU/l	35～75	*LAP
★GOT	19	21	27	21	IU/l	10～40	*GOT
★GPT	21	24	25	26	IU/l	6～40	*GPT
★LDH	291	310	286	293	IU/l	230～460	*LDH
★γ-GTP	17	21	23	23	IU/l	50以下	*γ-GTP
★総コレステロール	166	187	184	177	mg/dl	130～220	*総コレステロール
★トリグリセライド	79	105	123	98	mg/dl	35～150	*トリグリセライド
★尿素窒素	17	19	17	16	mg/dl	8～21	*尿素窒素
★クレアチニン	0.9	0.9	0.7	0.7	mg/dl	0.6～1.3	*クレアチニン
★尿酸	4.9	5.6	5.6	4.8	mg/dl	2.5～7.5	*尿酸
		5.6	79	91	mg/dl	70～110	*血糖
				5.3	%	4.3～5.8	*Hb-A1C
★白血球数	63						
★赤血球数	537	79	61				
★ヘモグロビン量	15.4	570	553	61	×10³/μl	36～92	*白血球数
★ヘマトクリット値	46.9	16.3	15.7	568	×10⁴/μl	420～560	*赤血球数
MCV	87	49.5	47.9	16.2	g/dl	13.0～17.0	*ヘモグロビン量
MCH	28.7	87	87	48.8	%	39.0～50.0	*ヘマトクリット値
MCHC	32.8	28.6	28.4	86	fl	81～98	MCV
★血小板数	22.6	32.9	32.8	28.5	pg	27.0～33.5	MCH
★血液像		23.1	22.9	33.2	%	32.0～35.0	MCHC
骨髄球	0.0			23.6	×10⁴/μl	14.0～35.0	*血小板数
後骨髄球	0.0	0.0	0.0				*血液像
好中球	52.3	0.0	0.0	0.0	%	0.0	骨髄球
好酸球	3.8	54.8	51.4	0.0	%	0.0	後骨髄球
好塩基球	1.0	3.9	4.6	51.9	%	36.0～69.0	好中球
リンパ球	37.6	1.0	1.3	4.4	%	1.0～5.0	好酸球
異型リンパ球	0.0	35.4	37.5	1.1	%	0.0～2.0	好塩基球
単球	5.3	0.0	0.0	35.4	%	27.0～53.0	リンパ球
大小不同		4.9	5.2	0.0	%	0.0	異型リンパ球
環状				7.2	%	2.0～10.0	単球
奇形							大小不同
赤芽球							環状
							奇形
							赤芽球

総合報告書I 受付 11/04/1 登録衛生検査 (株)メディッ TEL(059)2

受付11/04/27 登録衛生検査 (株)メディ TEL(059)2

受付11/05/11 登録衛生検査 (株)メディ TEL(059)2

受付11/05/25 報告11/05/26 登録衛生検査所 (株)メディック TEL(059)224-5455

総合報告書I

責任者 大谷 敦

[redacted]

責任者 大谷 敦

CONFIDENTIAL

[redacted]

検査成績報告書

クリニック 3389(2377) 津健康クリニック津健康クリニック

様 277
088

様 男性
67才

Male
K.S 67

コメント 混濁 (2+) 混濁 (1+)

定値		検査項目		測定値		測定値		単位	基準値
'03/02		採取		受付 11/03/16		11/03/30			報告 11/03/31
Serum total protein 8.1		★血清総蛋白	尿	7.6		7.4		g/dl	6.5～8.2
Albumin 1.3		★A/G比		1.2		1.4			1.1～2.0
4.6		★アルブミン		[illegible]		4.3		g/dl	3.7～5.3
4.8		★ZTT	尿	6.5		5.8		units	3.0～12.0
309		★ALP		279		273		IU/l	100～350
76		★LAP		65		58		IU/l	35～75
35		★GOT		22		25		IU/l	10～40
52		★GPT		29		33		IU/l	6～40
354		★LDH		333		320		IU/l	230～460
69		★γ-GTP	↗	53	↗	56		IU/l	50以下
Total Cholesterol 292		★総コレステロール	↑	270	↑	270		mg/dl	130～220
Triglyceride 374		★トリグリセライド	尿↑↑	580	尿↑	387		mg/dl	35～150
BUN 17		★尿素窒素		17		20		mg/dl	8～21
Creatinine 1.1		★クレアチニン		1.3		1.1		mg/dl	0.6～1.3
Uric acid 6.4		★尿酸		6.0		6.9		mg/dl	2.5～7.5
6.9		★Hb-A1C			↗	119		mg/dl	70～110
				62	↗	6.3		%	4.3～5.8
WBC 54		★白血球数		516					
RBC 518		★赤血球数		15.0		51		×10³/μl	36～92
Hemoglobin 15.2		★ヘモグロビン量		48.6		513		×10⁴/μl	420～560
Hematocrit 47.9		★ヘマトクリット値		94		15.4		g/dl	13.0～17.0
92		MCV		29.1		47.7		%	39.0～50.0
29.3		MCH	↓	30.9		93		fl	81～98
31.7		MCHC		24.7		30.0		pg	27.0～33.5
Platelet 25.7		★血小板数				32.3		%	32.0～35.0
				0.0		24.8		×10⁴/μl	14.0～35.0
				0.0					
				51.5		0.0		%	0.0
				1.4		0.0		%	0.0
				1.1		47.7		%	36.0～69.0
				42.6		2.2		%	1.0～5.0
				0.0		1.4		%	0.0～2.0
				3.4		45.7		%	27.0～53.0
						0.0		%	0.0
						3.0		%	2.0～10.0

/03/02 総合報告書Ⅰ 受付 11/03/1[illegible] 受付 11/03/30 報告 11/03/31

[illegible]録衛生検査所 登録衛生検査所 実施料 329点

ディッ 担当医 大谷 敦 (株)メディ・ (株)メディック

EL(059)22 TEL(059)2 TEL(059)224-5455

[redacted] 検査成績報告書

CONFIDENTIAL

検査成績報告書

津健康クリニック

3389(2377) 津健康クリニック [redacted] 様

[redacted]

男性 67才

実施料 329点 | 実施料 244点 | 実施料 329点

コメント 混濁(2+) | IDNo 2-77002 | IDNo 2-77083 | IDNo 2-77089 | コメント 混濁(2+)

採取 | 受付 11/04/13 | 受付 11/04/27 | 受付 11/05/11 | 採取 | 受付 11/05/25 | 報告 11/05/26

検査項目	測定値	測定値	測定値	検査項目	測定値	単位	基準値
★血清総蛋白	7.1	7.6	7.2	*血清総蛋白	7.2	g/dl	6.5～8.2
★A/G比	1.4	1.3	1.4	*A/G比	1.5		1.1～2.0
★アルブミン	4.2	4.3	4.2	*アルブミン	4.3	g/dl	3.7～5.3
★ZTT	5.3	4.3	5.0	*ZTT	4.0	units	3.0～12.0
★ALP	273	256	246	*ALP	283	IU/l	100～350
★LAP	56	58	52	*LAP	53	IU/l	35～75
★GOT	19	25	17	*GOT	23	IU/l	10～40
★GPT	25	28	23	*GPT	27	IU/l	6～40
★LDH	289	326	329	*LDH	345	IU/l	230～460
★γ-GTP	41	42	30	*γ-GTP	31	IU/l	50以下
★総コレステロール	243	268	227	*総コレステロール	238	mg/dl	130～220
★トリグリセライド	378	369	190	*トリグリセライド	374	mg/dl	35～150
★尿素窒素	21	18	18	*尿素窒素	19	mg/dl	8～21
★クレアチニン	1.1	1.1	1.1	*クレアチニン	0.9	mg/dl	0.6～1.3
★尿酸	6.5	5.9	4.9	*尿酸	5.8	mg/dl	2.5～7.5
★血糖空腹時	113	99	102	*血糖	109	mg/dl	70～110
		6.9		*Hb-AiC	6.3	%	4.3～5.8
★白血球数	64		83				
★赤血球数	513	51	518	*白血球数	52	×10⁴/μl	36～92
★ヘモグロビン量	15.6	530	15.7	*赤血球数	539	×10⁴/μl	420～560
★ヘマトクリット値	47.6	16.0	47.9	*ヘモグロビン量	16.3	g/dl	13.0～17.0
MCV	93	49.7	92	*ヘマトクリット値	49.3	%	39.0～50.0
MCH	30.4	94	30.3	MCV	91	fl	81～98
MCHC	32.8	30.2	32.8	MCH	30.2	pg	27.0～33.5
★血小板数	22.8	32.2	25.8	MCHC	33.1	%	32.0～35.0
★血液像		22.6		*血小板数	23.5	×10⁴/μl	14.0～35.0
骨髄球	0.0		0.0	*血液像			
後骨髄球	0.0	0.0	0.0	骨髄球	0.0	%	0.0
好中球	57.0	0.0	73.9	後骨髄球	0.0	%	0.0
好酸球	1.7	46.3	0.2	好中球	47.4	%	36.0～69.0
好塩基球	0.8	2.2	0.4	好酸球	3.3	%	1.0～5.0
リンパ球	36.4	1.2	23.0	好塩基球	1.3	%	0.0～2.0
異型リンパ球	0.0	46.6	0.0	リンパ球	44.9	%	27.0～53.0
単球	4.1	0.0	2.5	異型リンパ球	0.0	%	0.0
大小不同		3.7		単球	3.1	%	2.0～10.0
環状				大小不同			
奇形				環状			
赤芽球				奇形			
				赤芽球			

総合報告書Ⅰ 受付 11/04/1 | 受付 11/04/27 | 受付 11/05/11 | 総合報告書Ⅰ 受付 11/05/25 報告 11/05/26

登録衛生検査 | 登録衛生検査 | 登録衛生検査 | 登録衛生検査所

責任者 大谷 敦 | (株)メディ TEL(059)2 | (株)メディ TEL(059)2 | 責任者 大谷 敦 | (株)メディック TEL(059)224-5455

CONFIDENTIAL

総合報告書　総合報告書

3389(2377)　津健康クリニック　津健康クリニック　津健康クリニック

ID No. 277 082

[redacted]

様 男性 49才

Male
H.N [redacted] 49

採取		受付 11/03/02	受付 11/03/16	受付 11/03/30	報告 11/03/31		
	検査項目	測定値	測定値	測定値	単位	基準値
Serum total protein	★血清総蛋白	8.2	8.1	8.0	g/dl	6.5～8.2
Albumin	★A/G比	1.1	1.3	1.2		1.1～2.0
	★アルブミン	4.2	4.5	4.4	g/dl	3.7～5.3
	★ZTT	5.9	7.0	7.2	units	3.0～12.0
	★ALP	188	186	177	IU/l	100～350
	★LAP	64	67	60	IU/l	35～75
	★GOT	25	23	20	IU/l	10～40
	★GPT	29	27	19	IU/l	6～40
	★LDH	403	371	395	IU/l	230～460
	★γ-GTP	25	21	18	IU/l	50以下
Total cholesterol	★総コレステロール	⇈ 312	↑ 295	↑ 284	mg/dl	130～220
Triglyceride	★トリグリセライド	↑ 327	↗ 226	↗ 255	mg/dl	35～150
BUN	★尿素窒素	14	15	13	mg/dl	8～21
Creatinine	★クレアチニン	1.3	1.3	↗ 1.4	mg/dl	0.6～1.3
Uric acid	★尿酸	7.5	↗ 8.2	↗ 7.8	mg/dl	2.5～7.5
	★Hb-A1C	4.7		4.6	%	4.3～5.8
			48			
WBC	★白血球数	73	466	63	×10³/μl	36～92
RBC	★赤血球数	550	13.7	547	×10⁴/μl	420～560
Hemoglobin	★ヘモグロビン量	↗ 17.5	45.0	↗ 17.8	g/dl	13.0～17.1
Hematocrit	★ヘマトクリット値	↗ 52.2	97	↗ 52.7	%	39.0～50.1
	MCV	95	29.4	96	fl	81～98
	MCH	31.8	↓ 30.4	32.5	pg	27.0～33.1
	MCHC	33.5	19.3	33.8	%	32.0～35.1
Platelet	★血小板数	25.6		21.5	×10⁴/μl	14.0～35.1
			0.0			
			0.0	0.0	%	0.0
			57.0	0.0	%	0.0
			3.1	59.6	%	36.0～69.0
			0.6	2.5	%	1.0～5.0
			33.1	0.8	%	0.0～2.0
			0.0	32.5	%	27.0～53.0
			6.2	0.0	%	0.0
				4.6	%	2.0～10.0

総合報告書Ⅰ　受付 11/03/0　受付 11/03/1　受付 11/03/30　報告 11/03/31

登録衛生検査　登録衛生検査　登録衛生検査所　実施料 329点

責任者 大谷 敦

(株)メディック　(株)メディック　(株)メディック

TEL(059)2　TEL(059)2　TEL(059)224-5455

検査成績報告書

CONFIDENTIAL

検査成績報告書

3389(2377) 津健康クリニック [redacted]

実施料 329点 | コメント 混濁(1+) | IDNo 2-77085 | 受付 11/04/14

検査項目	測定値	測定値	測定値
★血清総蛋白	7.7	7.4	7.9
★A/G比	1.3	1.2	1.3
★アルブミン	4.3	4.0	4.4
★ZTT	7.5	7.3	7.2
★ALP	184	170	181
★LAP	60	59	62
★GOT	19	19	19
★GPT	21	22	17
★LDH	343	342	371
★γ-GTP	17	18	18
★総コレステロール	↑ 264	↑ 263	↑ 283
★トリグリセライド	(脂)↑ 358	(脂)↑ 371	↗ 304
★尿素窒素	16	14	15
★クレアチニン	↗ 1.5	↗ 1.4	↗ 1.4
★尿酸	↗ 7.7	↗ 8.6	↗ 8.2
		5.1	
★白血球数	71		72
★赤血球数	542	56	541
★ヘモグロビン量	↗ 17.6	528	↗ 17.4
★ヘマトクリット値	↗ 51.8	↗ 17.1	↗ 51.7
MCV	96	↗ 50.4	96
MCH	32.5	95	32.2
MCHC	34.0	32.4	33.7
★血小板数	21.8	33.9	21.0
★血液像		21.3	
骨髄球	0.0		0.0
後骨髄球	0.0	0.0	0.0
好中球	63.9	0.0	64.2
好酸球	3.9	59.8	3.5
好塩基球	0.8	4.8	1.0
リンパ球	27.3	0.7	26.4
異型リンパ球	0.0	29.4	0.0
単球	4.1	0.0	4.9
大小不同		5.3	
環状			
奇形			
赤芽球			

総合報告書Ⅰ

受付 11/04/14 | 受付 11/04/28 | 受付 11/05/12

登録衛生検査所 (株)メディッ TEL(059)22 | 登録衛生検査所 (株)メディ TEL(059)2 | 登録衛生検査所 (株)メディ TEL(059)2

責任者 大谷 敦

検査成績報告書

3389(2377) 津健康クリニック 様

[redacted] 様

男性 49才

実施料 329点 | コメント | IDNo 2-77087

採取 | 受付 11/05/26 | 報告 11/05/27

検査項目	測定値	単位	基準値
*血清総蛋白	7.9	g/dℓ	6.5～8.2
*A/G比	1.2		1.1～2.0
*アルブミン	4.3	g/dℓ	3.7～5.3
*ZTT	7.3	units	3.0～12.0
*ALP	178	IU/ℓ	100～350
*LAP	61	IU/ℓ	35～75
*GOT	18	IU/ℓ	10～40
*GPT	18	IU/ℓ	6～40
*LDH	366	IU/ℓ	230～460
*γ-GTP	17	IU/ℓ	50以下
*総コレステロール	↑ 285	mg/dℓ	130～220
*トリグリセライド	↑ 332	mg/dℓ	35～150
*尿素窒素	16	mg/dℓ	8～21
*クレアチニン	1.2	mg/dℓ	0.6～1.3
*尿酸	↗ 8.1	mg/dℓ	2.5～7.5
*Hb-AIC	4.8	%	4.3～5.8
*白血球数	67	×10³/μℓ	36～92
*赤血球数	541	×10⁴/μℓ	420～560
*ヘモグロビン量	↗ 17.4	g/dℓ	13.0～17.0
*ヘマトクリット値	↗ 51.0	%	39.0～50.0
MCV	94	fℓ	81～98
MCH	32.2	Pg	27.0～33.5
MCHC	34.1	%	32.0～35.0
*血小板数	21.9	×10⁴/μℓ	14.0～35.0
*血液像			
骨髄球	0.0	%	0.0
後骨髄球	0.0	%	0.0
好中球	56.5	%	36.0～69.0
好酸球	3.9	%	1.0～5.0
好塩基球	1.0	%	0.0～2.0
リンパ球	34.6	%	27.0～53.0
異型リンパ球	0.0	%	0.0
単球	4.0	%	2.0～10.0
大小不同			
環状			
奇形			
赤芽球			

総合報告書Ⅰ

受付 11/05/26 | 報告 11/05/27

登録衛生検査所 (株)メディック TEL(059)224-5455

責任者 大谷 敦

CONFIDENTIAL

[redacted]

[redacted] 告書 告書

3389(2377) 津健康クリニック 津健康クリニック 津健康クリニック

ID No. 277 081

様 男性 34才 Male Y.I 34

[redacted]

検査項目	測定値	測定値	測定値	単位	基準値
採取	受付 11/03/02	受付 11/03/16	受付 11/03/30	報告 11/03/31	
★血清総蛋白	8.1	7.8	8.0	g/dl	6.5～8.2
★A/G比	1.3	1.4	1.3		1.1～2.0
★アルブミン	4.6	4.5	4.5	g/dl	3.7～5.3
★ZTT	7.3	8.8	9.0	units	3.0～12.0
★ALP	246	248	247	IU/l	100～350
★LAP	↗ 85	↗ 81	↗ 76	IU/l	35～75
★GOT	26	20	17	IU/l	10～40
★GPT	↗ 41	33	26	IU/l	6～40
★LDH	366	429	391	IU/l	230～460
★γ-GTP	30	29	26	IU/l	50以下
★総コレステロール	↗ 223	202	220	mg/dl	130～220
★トリグリセライド	125	↗ 157	73	mg/dl	35～150
★尿素窒素	14	15	21	mg/dl	8～21
★クレアチニン	0.8	0.9	0.8	mg/dl	0.6～1.3
★尿酸	5.0	5.8	5.2	mg/dl	2.5～7.5
★Hb-AIC	4.9		4.7	%	4.3～5.8
		65			
★白血球数	56	483	76	×10³/μl	36～92
★赤血球数	542	13.1	543	×10⁴/μl	420～560
★ヘモグロビン量	15.1	42.8	15.5	g/dl	13.0～17.
★ヘマトクリット値	46.8	89	47.8	%	39.0～50.
MCV	86	27.1	88	fl	81～98
MCH	27.9	↓ 30.6	28.5	pg	27.0～33.
MCHC	32.3	23.2	32.4	%	32.0～35.
★血小板数	23.0		23.8	×10⁴/μl	14.0～35.
		0.0			
		0.0	0.0	%	0.0
		52.3	0.0	%	0.0
		↗ 6.3	60.9	%	36.0～69.
		0.3	↑ 11.1	%	1.0～5.0
		36.1	0.8	%	0.0～2.0
		0.0	↘ 24.2	%	27.0～53.
		5.0	0.0	%	0.0
			3.0	%	2.0～10.

総合報告書Ⅰ

受付 11/03/0 登録衛生検査 (株)メディッ TEL(059)2

受付 11/03/1 登録衛生検査 (株)メディッ TEL(059)2

受付 11/03/30 登録衛生検査所 (株)メディック TEL(059)224-5455

報告 11/03/3 実施料 329

担当者 大谷 敦

検査成績報告

CONFIDENTIAL

検査成績報告書 検査成績報告 検査成績報告 検査成績報告書

津健康クリニック 津健康クリニック 津健康クリニック 様

3389(2377) 津健康クリニック [redacted] 様

男性 34才

		実施料 329点	実施料 244点	実施料 329点			
	コメント	IDNo 2-77001	IDNo 2-77081	IDNo 2-77091			乳腺 コメント
採取	受付	11/04/13 受付	11/04/27 受付	11/05/11 受付	11/05/25		報告 11/05/26
検査項目	測定値	測定値	測定値	測定値		単位	基準値
★血清総蛋白	7.8	7.7	7.5	8.0	高	g/dl	6.5～8.2
★A/G比	1.4	1.2	1.3	1.2			1.1～2.0
★アルブミン	4.5	4.2	4.2	4.3		g/dl	3.7～5.3
★ZTT	9.7	9.6	9.8	10.8	高	units	3.0～12.0
★ALP	232	238	234	243		IU/l	100～350
★LAP	↗ 81	75	↗ 80	73		IU/l	35～75
★GOT	21	22	20	16		IU/l	10～40
★GPT	39	38	36	22		IU/l	6～40
★LDH	356	370	377	385		IU/l	230～460
★γ-GTP	28	32	35	26		IU/l	50以下
★総コレステロール	202	204	211	202		mg/dl	130～220
★トリグリセライド	111	↗ 159	139	428	高	mg/dl	35～150
★尿素窒素	19	21	↗ 25	14		mg/dl	8～21
★クレアチニン	0.9	0.9	0.9	0.8		mg/dl	0.6～1.3
★尿酸	5.7	5.2	6.5	5.8		mg/dl	2.5～7.5
		5.3		4.8		%	4.3～5.8
★白血球数	65		74				
★赤血球数	556	67	504	72		×10⁴/μl	36～92
★ヘモグロビン量	15.7	525	14.3	523		×10⁴/μl	420～560
★ヘマトクリット値	48.4	15.0	43.8	15.0		g/dl	13.0～17.0
MCV	87	45.9	87	45.1		%	39.0～50.0
MCH	28.2	87	28.4	86		fl	81～98
MCHC	32.4	28.6	32.6	28.7		pg	27.0～33.5
★血小板数	23.3	32.7	22.3	33.3		%	32.0～35.0
★血液像		22.7		25.7		×10⁴/μl	14.0～35.0
骨髄球	0.0		0.0				
後骨髄球	0.0	0.0	0.0	0.0		%	0.0
好中球	54.0	0.0	64.8	0.0		%	0.0
好酸球	↗ 9.4	61.4	4.6	62.3		%	36.0～69.0
好塩基球	0.8	↗ 5.4	0.5	4.7		%	1.0～5.0
リンパ球	31.3	0.8	27.7	0.4		%	0.0～2.0
異型リンパ球	0.0	27.6	0.0	28.7		%	27.0～53.0
単球	4.5	0.0	2.4	0.0		%	0.0
大小不同		4.8		3.9		%	2.0～10.0
環状							
奇形							
赤芽球							

総合報告書Ⅰ 採取 11/04/1 受付 11/04/27 受付 11/05/11 受付 11/05/25 報告 11/05/26

	登録衛生検査所	登録衛生検査	登録衛生検査	登録衛生検査所
責任者	㈱メディッ	㈱メディ	㈱メディ	㈱メディック
大谷 敦	TEL(059)22	TEL(059)2	TEL(059)22	TEL(059)224-5455

CONFIDENTIAL

検査成績報告書

3389(2377)　津健康クリニック　津健康クリニック　津健康クリニック

IDNo 290 350

[redacted]

様 女性

Hemal
A.M 33

採取		コメント	受付 11/03/02	コメント	受付 11/03/16	コメント	受付 11/03/30	報告 11/03/31	
	検査項目		測定値		測定値		測定値	単位	基準値
Serum total protein	★血清総蛋白		7.7		7.6		7.4	g/dℓ	6.5～8.2
	★A/G比		1.3		1.3		1.4		1.1～2.0
Albumin	★アルブミン		4.3		4.3		4.3	g/dℓ	3.7～5.3
	★ZTT		7.6		11.0		9.8	units	3.0～12.0
	★ALP		188		170		176	IU/ℓ	100～350
	★LAP		52		57		49	IU/ℓ	35～75
	★GOT		14		15		11	IU/ℓ	10～40
	★GPT		12		16		11	IU/ℓ	6～40
	★LDH		268		262		264	IU/ℓ	230～460
	★γ-GTP		15		14		14	IU/ℓ	50以下
Total cholesterol	★総コレステロール	↗	221	↗	236		178	mg/dℓ	130～220
Triglyceride	★トリグリセライド		80		135		130	mg/dℓ	35～150
BUN	★尿素窒素		15		15		13	mg/dℓ	8～21
Creatinine	★クレアチニン		0.9		1.0		0.9	mg/dℓ	0.6～1.3
Uric acid	★尿酸		5.6		5.2		5.3	mg/dℓ	2.0～6.5
	★Hb-AlC		4.5				4.5	%	4.3～5.8
					59				
WBC	★白血球数		56		413		57	×10³/μℓ	36～92
RBC	★赤血球数	↗	513		11.9		491	×10⁴/μℓ	380～500
Hemoglobin	★ヘモグロビン量	↗	14.9		38.7		14.1	g/dℓ	11.2～14.7
Hematocrit	★ヘマトクリット値	↗	46.8		94	↗	44.7	%	33.5～43.5
	MCV		91		28.8		91	fℓ	81～98
	MCH		29.0	↓	30.7		28.7	pg	27.0～33.5
	MCHC	↘	31.8		24.4	↘	31.5	%	32.0～35.0
Platelet	★血小板数		32.7				31.4	×10⁴/μℓ	14.0～35.0
					0.0				
					0.0		0.0	%	0.0
					57.9		0.0	%	0.0
				↗	5.4		46.2	%	36.0～69.0
					1.0	↗	5.7	%	1.0～5.0
					29.0		0.7	%	0.0～2.0
					0.0		40.8	%	27.0～53.0
					6.7		0.0	%	0.0
							6.6	%	2.0～10.0

総合報告書Ⅰ　受付 11/03/0　受付 11/03/1　受付 11/03/30　報告 11/03/31

登録衛生検査　登録衛生検査　登録衛生検査所　実施料 329点

担任者 大谷 敦

(株)メディ・(株)メディ・(株)メディック

TEL(059)2　TEL(059)2　TEL(059)224-5455

検査成績報告書

検査成績報告書 検査成績報告書 検査成績報告書

389(2377) 沖健康クリニック [redacted]

沖健康クリニック [redacted]

沖健康クリニック [redacted]

様

CONFIDENTIAL

女性 33才

実施料 329点	実施料 244点	実施料 329点	コメント
ID№ 2-77086	ID№ 2-90351	ID№ 2-77088	
受付11/04/28	受付11/05/12	受付11/05/28	報告11/05/29

		前々回	前回		単位	基準値
11/04/13						
血清総蛋白	7.6	7.4	7.3	7.5	g/dl	6.5~8.2
A/G比	1.3	1.2	1.3	1.2		1.1~2.0
アルブミン	4.3	4.1	4.1	4.1	g/dl	3.7~5.3
ZTT	8.9	9.5	8.7	9.7	units	3.0~12.0
ALP	169	156	169	172	IU/l	100~350
LAP	54	49	52	54	IU/l	35~75
GOT	15	15	16	16	IU/l	10~40
GPT	15	16	17	19	IU/l	6~40
LDH	293	278	290	257	IU/l	230~460
γ-GTP	14	16	14	12	IU/l	50以下
総コレステロール	193	191	189	179	mg/dl	130~220
トリグリセライド	109	95	95	95	mg/dl	35~150
尿素窒素	18	14	14	11	mg/dl	8~21
クレアチニン	1.1	1.0	1.0	1.0	mg/dl	0.6~1.3
尿酸	6.4	5.7	5.0	4.8	mg/dl	2.0~6.5
		5.0		4.4	%	4.3~5.8
白血球数	↗ 94		75			
赤血球数	480	46	473	52	×10⁴/μl	36~92
ヘモグロビン量	↗ 14.8	468	14.3	468	×10⁴/μl	380~500
ヘマトクリット値	43.5	14.2	43.4	14.1	g/dl	11.2~14.7
MCV	91	42.6	92	42.5	%	33.5~43.5
MCH	30.8	91	30.2	91	fl	81~98
MCHC	34.0	30.3	32.9	30.1	pg	27.0~33.5
血小板数	28.8	33.3	22.8	33.2	%	32.0~35.0
血液像		28.3		24.3	×10⁴/μl	14.0~35.0
骨髄球	0.0		0.0			
後骨髄球	0.0	0.0	0.0	0.0	%	0.0
桿中球	59.3	0.0	61.8	0.0	%	0.0
桿酸球	↗ 5.2	45.2	↗ 5.1	52.4	%	36.0~69.0
好塩基球	0.5	↗ 6.8	0.1	↗ 5.2	%	1.0~5.0
リンパ球	29.8	1.1	27.1	1.0	%	0.0~2.0
異型リンパ球	0.0	41.2	0.0	37.7	%	27.0~53.0
単球	5.2	0.0	5.9	0.0	%	0.0
大小不同		5.7		3.7	%	2.0~10.0
腫状						
奇形						
赤芽球						

総合報告書1

11/04/1	受付11/04/28	受付11/05/12	受付11/05/28 報告11/05/29
登録衛生検査	登録衛生検査	登録衛生検査	登録衛生検査所
(株)メディ	(株)メディ	(株)メディ	(株)メディック
TEL(0591	TEL(0591	TEL(0591	TEL(059)224-5455

[redacted]

CONFIDENTIAL

検査成績報告書

3389(2377)　津健康クリニック　津健康クリニック　津健康クリニック

[redacted]

ID NO 277 091

様 女性 65才

Femal K.S 65

採取		コメント	受付 11/03/03	コメント	受付 11/03/17	コメント	受付 11/04/01	報告 11/04/02	
	検査項目		測定値		測定値		測定値	単位	基準値
Serum total protein	★血清総蛋白		7.4		7.7		7.4	g/dl	6.5～8.2
	★A/G比		1.6		1.5		1.6		1.1～2.0
Albumin	★アルブミン		4.5		4.6		4.5	g/dl	3.7～5.3
	★ZTT		6.6		6.1		6.6	units	3.0～12.0
	★ALP	↗	374	↗	417	↗	360	IU/l	100～350
	★LAP		56		55		52	IU/l	35～75
	★GOT		23		20		25	IU/l	10～40
	★GPT		19		15		17	IU/l	6～40
	★LDH		357		317		372	IU/l	230～460
	★γ-GTP		14		14		12	IU/l	50以下
Total cholesterol	★総コレステロール		209		214		173	mg/dl	130～220
Triglyceride	★トリグリセライド	↗	206	↗	193		72	mg/dl	35～150
BUN	★尿素窒素		11		15		17	mg/dl	8～21
Creatinine	★クレアチニン	↘	0.5	↓	0.4	↘	0.5	mg/dl	0.6～1.3
Uric acid	★尿酸		4.9		5.4		4.6	mg/dl	2.0～6.5
	★Hb-A1C		5.5				5.4	%	4.3～5.8
					53				
WBC	★白血球数		50		442		43	×10³/μl	36～92
RBC	★赤血球数		426		13.6		407	×10⁴/μl	380～500
Hemoglobin	★ヘモグロビン量		12.9		42.4		12.6	g/dl	11.2～14.7
Hematocrit	★ヘマトクリット値		40.4		96	P	39.8	%	33.5～43.5
	MCV		95		30.8		98	fl	81～98
	MCH		30.3		32.1		31.0	pg	27.0～33.5
	MCHC	↘	31.9		27.1	↘	31.7	%	32.0～35.0
Platelet	★血小板数		26.2				24.7	×10⁴/μl	14.0～35.0
					0.0				
					0.0		0.0	%	0.0
					63.9		0.0	%	0.0
					1.9		43.9	%	36.0～69.0
					0.2		1.6	%	1.0～5.0
					26.3		0.7	%	0.0～2.0
					0.0		49.3	%	27.0～53.0
					7.7		0.0	%	0.0
							4.5	%	2.0～10.0

総合報告書Ⅰ　受付 11/03/0　受付 11/03/1　受付 11/04/01　報告 11/04/02

登録衛生検査　登録衛生検査　登録衛生検査所　実施料 329点

責任者 大谷 教

(株)メディ…　(株)メディ…　(株)メディック

TEL(059)2　TEL(059)2　TEL(059)224-5455

検査成績報告書

検査成績報告書

津健康クリニック　津健康クリニック　3389(2377)　津健康クリニック　様

[redacted]　[redacted]　様

女性　65才

検査項目	測定値	測定値	測定値	検査項目	測定値	単位	基準値
	実施料 329点	実施料 244点			実施料 329点		
	IDNo. 2-77082	IDNo. 2-77085			IDNo. 2-77084		
採取	受付 11/04/14	受付 11/04/28	受付 11/05/11	採取	受付 11/05/26		報告 11/05/27
★血清総蛋白	8.1	8.1	7.2	*血清総蛋白	7.6	g/dℓ	6.5～8.2
★A/G比	1.3	1.4	1.5	*A/G比	1.5		1.1～2.0
★アルブミン	4.6	4.7	4.3	*アルブミン	4.5	g/dℓ	3.7～5.3
★ZTT	6.6	6.6	6.4	*ZTT	5.4	units	3.0～12.0
★ALP	↗ 410	↗ 393	348	*ALP	↗ 368	IU/ℓ	100～350
★LAP	57	57	54	*LAP	56	IU/ℓ	35～75
★GOT	25	23	23	*GOT	25	IU/ℓ	10～40
★GPT	20	19	19	*GPT	20	IU/ℓ	6～40
★LDH	354	323	300	*LDH	393	IU/ℓ	230～460
★γ-GTP	14	15	26	*γ-GTP	10	IU/ℓ	50以下
★総コレステロール	↗ 228	↗ 222	216	*総コレステロール	185	mg/dℓ	130～220
★トリグリセライド	↗ 195	↗ 207	↗ 197	*トリグリセライド	150	mg/dℓ	35～150
★尿素窒素	17	14	13	*尿素窒素	17	mg/dℓ	8～21
★クレアチニン	0.6	0.6	0.6	*クレアチニン	↘ 0.5	mg/dℓ	0.6～1.3
★尿酸	5.4	5.7	4.8	*尿酸	4.6	mg/dℓ	2.0～6.5
		5.3		*Hb-A1C	5.2	%	4.3～5.8
★白血球数	43		37				
★赤血球数	451	40	422	*白血球数	39	×10²/μℓ	36～92
★ヘモグロビン量	13.9	450	13.3	*赤血球数	440	×10⁴/μℓ	380～500
★ヘマトクリット値	43.0	14.1	39.8	*ヘモグロビン量	13.7	g/dℓ	11.2～14.7
MCV	95	42.4	94	*ヘマトクリット値	42.4	%	33.5～43.5
MCH	30.8	94	31.5	MCV	96	fℓ	81～98
MCHC	32.3	31.3	33.4	MCH	31.1	pg	27.0～33.5
★血小板数	26.1	33.3	24.4	MCHC	32.3	%	32.0～35.0
★血液像		25.8		*血小板数	26.5	×10⁴/μℓ	14.0～35.0
骨髄球	0.0		0.0	*血液像			
後骨髄球	0.0	0.0	0.0	骨髄球	0.0	%	0.0
桿状核		0.0	↘ 33.6	後骨髄球	0.0	%	0.0
分葉核		42.9	2.5	好中球	36.6	%	36.0～69.0
好酸球	2.3	2.5	0.5	好酸球	3.4	%	1.0～5.0
好塩基球	0.7	0.2	↑ 60.9	好塩基球	0.3	%	0.0～2.0
リンパ球	↗ 58.1	52.4	0.0	リンパ球	↗ 55.3	%	27.0～53.0
異型リンパ球	0.0	0.0	2.5	異型リンパ球	0.0	%	0.0
単球	2.1	2.0		単球	4.4	%	2.0～10.0
大小不同				大小不同			
環状				環状			
奇形				奇形			
赤芽球				赤芽球			
総合報告書Ⅰ	受付 11/04/1	受付 11/04/28	受付 11/05/11	総合報告書Ⅰ	受付 11/05/26		報告 11/05/27
	登録衛生検査	登録衛生検査	登録衛生検査		登録衛生検査所		
責任者 大谷 敦	㈱メディ TEL(059)2	㈱メディ TEL(059)2	㈱メディ TEL(059)2	責任者 大谷 敦	㈱メディック TEL(059)224-5455		

[redacted]

CONFIDENTIAL

検査成績報告書

3389(2377)　津健康クリニック　津健康クリニック　津健康クリニック

様 277 073

様 女性 42才

Fremal S.M 42

採取		コメント	受付 11/03/03	コメント	11/03/17	コメント	受付 11/03/31	報告 11/04/01	
	検査項目		測定値		測定値		測定値	単位	基準値
Serum total protein	★血清総蛋白		7.4		7.5		7.6	g/dl	6.5～8.2
	★A/G比		1.3		1.3		1.3		1.1～2.0
Albumin	★アルブミン		4.2		4.3		4.3	g/dl	3.7～5.3
	★ZTT		4.6		6.0		5.1	units	3.0～12.0
	★ALP		213		203		174	IU/l	100～350
	★LAP		45		47		46	IU/l	35～75
	★GOT		18		20		20	IU/l	10～40
	★GPT		16		23		21	IU/l	6～40
	★LDH		279		274		277	IU/l	230～460
	★γ-GTP		14		17		15	IU/l	50以下
Total cholestrol	★総コレステロール		215		181		207	mg/dl	130～220
Triglyceride	★トリグリセライド	↗	199	↗	156	↗	201	mg/dl	35～150
BUN	★尿素窒素		12		14		16	mg/dl	8～21
Creatinine	★クレアチニン		0.7		0.8		0.7	mg/dl	0.6～1.3
Uric acid	★尿酸		3.4		4.2		4.2	mg/dl	2.0～6.5
	★Hb-A1C		5.0				4.8	%	4.3～5.8
					71				
WBC	★白血球数		59		425		70	×10³/μl	36～92
RBC	★赤血球数		426		12.8		416	×10⁴/μl	380～500
Hemoglobin	★ヘモグロビン量		13.1		40.9		13.0	g/dl	11.2～14.7
Hematocrit	★ヘマトクリット値		40.3		96		40.2	%	33.5～43.5
	MCV		95		30.1		97	fl	81～98
	MCH		30.8	↘	31.3		31.3	pg	27.0～33.5
	MCHC		32.5		24.4		32.3	%	32.0～35.0
Platelet	★血小板数		28.1				28.2	×10⁴/μl	14.0～35.0
					0.0				
					0.0		0.0	%	0.0
					61.6		0.0	%	0.0
					2.2		54.4	%	36.0～69.0
					0.6		4.6	%	1.0～5.0
					30.8		1.0	%	0.0～2.0
					0.0		35.6	%	27.0～53.0
					4.8		0.0	%	0.0
							4.4	%	2.0～10.0

総合報告書Ⅰ　受付 11/03/0　受付 11/03/1　受付 11/03/31　報告 11/04/01

登録衛生検査所　登録衛生検査所　登録衛生検査所　実施料 329点

責任者 大谷 敦

(株)メディッ　(株)メディッ　(株)メディック

TEL(059)2[illegible]　TEL(059)2[illegible]　TEL(059)224-5455

検査成績報告書

検査成績報告書

津健康クリニック　津健康クリニック　3389(2377)　津健康クリニック　様

3389(2377)　津健康クリニック　[redacted]　様

[redacted]　女性　42才

検査項目	測定値	測定値	測定値	検査項目	測定値	単位	基準値
	実施料 329点	実施料 244点			実施料 329点		
採取	コメント	IDNo. 2-77083	IDNo. 2-77083	採取	IDNo. 2-77088	コメント	
	受付 11/04/14	受付 11/04/28	受付 11/05/12		受付 11/05/26	報告 11/05/27	
★血清総蛋白	7.3	7.2	7.0	*血清総蛋白	7.3	g/dl	6.5～8.2
★A/G比	1.4	1.3	1.3	*A/G比	1.3		1.1～2.0
★アルブミン	4.2	4.1	4.0	*アルブミン	4.1	g/dl	3.7～5.3
★ZTT	5.6	5.1	5.5	*ZTT	4.9	units	3.0～12.0
★ALP	159	194	187	*ALP	157	IU/l	100～350
★LAP	43	45	43	*LAP	43	IU/l	35～75
★GOT	22	21	17	*GOT	16	IU/l	10～40
★GPT	17	24	20	*GPT	13	IU/l	6～40
★LDH	322	336	296	*LDH	309	IU/l	230～460
★γ-GTP	14	13	16	*γ-GTP	12	IU/l	50以下
★総コレステロール	166	183	189	*総コレステロール	194	mg/dl	130～220
★トリグリセライド	113	89	96	*トリグリセライド	83	mg/dl	35～150
★尿素窒素	12	19	17	*尿素窒素	16	mg/dl	8～21
★クレアチニン	0.7	0.8	0.7	*クレアチニン	0.7	mg/dl	0.6～1.3
★尿酸	4.0	3.7	3.7	*尿酸	3.9	mg/dl	2.0～6.5
		5.2		*Hb-AlC	4.8	%	4.3～5.8
★白血球数	79		75				
★赤血球数	412	69	417	*白血球数	63	×10³/μl	36～92
★ヘモグロビン量	13.0	400	13.0	*赤血球数	409	×10⁴/μl	380～500
★ヘマトクリット値	39.2	12.8	40.0	*ヘモグロビン量	12.7	g/dl	11.2～14.7
MCV	95	37.2	96	*ヘマトクリット値	38.8	%	33.5～43.5
MCH	31.6	93	31.2	MCV	95	fl	81～98
MCHC	33.2	32.0	32.5	MCH	31.1	pg	27.0～33.5
★血小板数	23.5	34.4	25.6	MCHC	32.7	%	32.0～35.0
★血液像		24.8		*血小板数	24.7	×10⁴/μl	14.0～35.0
骨髄球	0.0		0.0	*血液像			
後骨髄球	0.0	0.0	0.0	骨髄球	0.0	%	0.0
桿状核		0.0	56.5	後骨髄球	0.0	%	0.0
分葉核		4.0	9.3	好中球	45.5	%	36.0～69.0
好酸球	↑ 13.5	42.0	1.1	好酸球	↑ 10.6	%	1.0～5.0
好塩基球	0.9	↑ 11.0	29.0	好塩基球	0.8	%	0.0～2.0
リンパ球	30.5	1.0	0.0	リンパ球	37.0	%	27.0～53.0
異型リンパ球	0.0	39.0	4.1	異型リンパ球	0.0	%	0.0
単球	5.2	0.0		単球	6.1	%	2.0～10.0
大小不同		3.0		大小不同			
環状				環状			
奇形				奇形			
赤芽球				赤芽球			

総合報告書Ⅰ　受付 11/04/1　受付 11/04/28　受付 11/05/12　総合報告書Ⅰ　受付 11/05/26　報告 11/05/27

登録衛生検査　登録衛生検査　登録衛生検査　登録衛生検査所

責任者 大谷 敦　(株)メディ　TEL(059)2　(株)メディ　TEL(059)2　(株)メディ　TEL(059)2　責任者 大谷 敦　(株)メディック　TEL(059)224-5455

[redacted]

CONFIDENTIAL

検査成績報告書 告書

｜クリニック 3389(2377) 津健康クリニック 津健康クリニック ID No. 277 076

[redacted] [redacted]

女性 29才

Female E.S 29

コメント 混濁 (1+) コメント 混濁 (1+)

	03/03	採取	11/03/1	受付 11/03/31	報告 11/04/01	
	測定値	検査項目	測定値	測定値	単位	基準値
Serum total protein	7.3	★血清総蛋白	7.7	7.8	g/dl	6.5~8.2
	1.1	★A/G比	1.1	1.2		1.1~2.0
Albumin	3.8	★アルブミン	4.1	4.2	g/dl	3.7~5.3
	9.5	★ZTT	↗ 12.2	↗ 13.7	units	3.0~12.0
	144	★ALP	151	156	IU/l	100~350
	52	★LAP	53	52	IU/l	35~75
	17	★GOT	12	15	IU/l	10~40
	7	★GPT	7	9	IU/l	6~40
	499	★LDH	321	301	IU/l	230~460
	8	★γ-GTP	8	7	IU/l	50以下
Total cholesterol	161	★総コレステロール	175	177	mg/dl	130~220
Triglyceride	72	★トリグリセライド	71	85	mg/dl	35~150
BUN	8	★尿素窒素	14	13	mg/dl	8~21
Creatinine	0.7	★クレアチニン	0.7	0.6	mg/dl	0.6~1.3
Uric acid	3.6	★尿酸	4.1	4.4	mg/dl	2.0~6.5
	5.2	★Hb-A1C		4.7	%	4.3~5.8
			79			
WBC	45	★白血球数	469	75	$\times10^2/\mu l$	36~92
RBC	496	★赤血球数	12.5	495	$\times10^4/\mu l$	380~500
Hemoglobin	3.1	★ヘモグロビン量	39.3	13.5	g/dl	11.2~14.7
Hematocrit	41.2	★ヘマトクリット値	84	42.2	%	33.5~43.5
	83	MCV	↘ 26.7	85	fl	81~98
	26.4	MCH	↘ 31.8	27.3	pg	27.0~33.5
	31.8	MCHC	↗ 43.3	32.0	%	32.0~35.0
Platelet	14.2	★血小板数		↗ 35.9	$\times10^4/\mu l$	14.0~35.0
			0.0			
			0.0	0.0	%	0.0
			63.7	0.0	%	0.0
			2.9	58.1	%	36.0~69.0
			0.5	3.5	%	1.0~5.0
			↘ 25.7	0.5	%	0.0~2.0
			0.0	31.9	%	27.0~53.0
			7.2	0.0	%	0.0
				6.0	%	2.0~10.0

03/0 総合報告書 I 受付 11/03/1 受付 11/03/31 報告 11/04/01

録衛生検査 登録衛生検査 登録衛生検査所 実施料 329点

ディ 責任者 (株)メディ (株)メディック

L(059): 大谷 数 TEL(059)2 TEL(059)224-5455

検査成績報告書

CONFIDENTIAL

検査成績報告書

3389(2377) 津健康クリニック [redacted] 様

女性 29才

実施料 329点 IDNo. 2-77084 受付 11/04/14

検査項目	測定値
★血清総蛋白	7.5
★A/G比	1.3
★アルブミン	4.2
★ZTT	↗ 13.0
★ALP	138
★LAP	55
★GOT	14
★GPT	6
★LDH	311
★γ-GTP	7
★総コレステロール	170
★トリグリセライド	82
★尿素窒素	16
★クレアチニン	0.7
★尿酸	4.3
★白血球数	68
★赤血球数	483
★ヘモグロビン量	13.2
★ヘマトクリット値	39.7
MCV	82
MCH	27.3
MCHC	33.2
★血小板数	30.7
★血液像	
骨髄球	0.0
後骨髄球	0.0
桿状核	
分葉核	
好酸球	2.6
好塩基球	0.3
リンパ球	↓ 17.8
異型リンパ球	0.0
単球	6.9
大小不同	
環状	
奇形	
赤芽球	

総合報告書Ⅰ

受付 11/04/14

登録衛生検査

(株)メディ

TEL(059)2

責任者 大谷 敦

検査成績報告

津健康クリニック

実施料 244点 IDNo. 2-77085 受付 11/05/12

測定値
7.4
1.2
4.0
↗ 13.0
141
51
16
9
336
9
171
59
15
0.6
4.0
92
484
13.2
40.8
84
27.3
32.4
31.7
0.0
0.0
61.6
3.5
0.3
28.8
0.0
5.8

総合報告書Ⅰ

受付 11/05/12

登録衛生検査

(株)メディ

TEL(059)2

責任者

検査成績報告書

3389(2377) 津健康クリニック 様

[redacted] 様

女性 29才

実施料 329点 IDNo. 2-77085 受付 11/05/26 報告 11/05/27

コメント

検査項目	測定値	単位	基準値
*血清総蛋白	7.5	g/dl	6.5～8.2
*A/G比	1.1		1.1～2.0
*アルブミン	4.0	g/dl	3.7～5.3
*ZTT	11.8	units	3.0～12.0
*ALP	143	IU/l	100～350
*LAP	54	IU/l	35～75
*GOT	14	IU/l	10～40
*GPT	9	IU/l	6～40
*LDH	339	IU/l	230～460
*γ-GTP	7	IU/l	50以下
*総コレステロール	174	mg/dl	130～220
*トリグリセライド	93	mg/dl	35～150
*尿素窒素	14	mg/dl	8～21
*クレアチニン	0.6	mg/dl	0.6～1.3
*尿酸	4.5	mg/dl	2.0～6.5
*Hb-A1C	4.8	%	4.3～5.8
*白血球数	61	×10³/μl	36～92
*赤血球数	484	×10⁴/μl	380～500
*ヘモグロビン量	13.2	g/dl	11.2～14.7
*ヘマトクリット値	40.5	%	33.5～43.5
MCV	84	fl	81～98
MCH	27.3	pg	27.0～33.5
MCHC	32.6	%	32.0～35.0
*血小板数	↗ 43.2	×10⁴/μl	14.0～35.0
*血液像			
骨髄球	0.0	%	0.0
後骨髄球	0.0	%	0.0
好中球	61.1	%	36.0～69.0
好酸球	4.1	%	1.0～5.0
好塩基球	0.5	%	0.0～2.0
リンパ球	31.0	%	27.0～53.0
異型リンパ球	0.0	%	0.0
単球	3.3	%	2.0～10.0
大小不同			
環状			
奇形			
赤芽球			

総合報告書Ⅰ

受付 11/05/26 報告 11/05/27

登録衛生検査所

(株)メディック

TEL(059)224-5455

責任者 大谷 敦

[redacted]

検査成績報告書

3389(2377)	津健康クリニック	津健康クリニック	津健康クリニック
	[redacted]	[redacted]	[redacted]
コメント			
採取	11/03/02	11/03/16	11/03/30
受付	11/03/02	11/03/16	11/03/30
報告			11/03/31

IDNo. 277086
様 女性 65才
Renal S.K 65

検査項目	測定値	測定値	測定値	単位	基準値
★血清総蛋白	8.1	7.5	8.1	g/dl	6.5～8.2
★A/G比	1.4	1.3	1.3		1.1～2.0
★アルブミン	4.7	4.3	4.5	g/dl	3.7～5.3
★ZTT	↘ 2.7	4.1	4.3	units	3.0～12.
★ALP	203	202	191	IU/l	100～350
★LAP	50	51	46	IU/l	35～75
★GOT	20	19	18	IU/l	10～40
★GPT	11	9	7	IU/l	6～40
★LDH	285	266	263	IU/l	230～460
★γ-GTP	15	11	10	IU/l	50以下
★総コレステロール	↑ 260	220	↑ 272	mg/dl	130～220
★トリグリセライド	↗ 193	114	↗ 179	mg/dl	35～150
★尿素窒素	15	16	16	mg/dl	8～21
★クレアチニン	0.8	0.9	0.7	mg/dl	0.6～1.3
★尿酸	3.9	4.0	4.4	mg/dl	2.0～6.5
★Hb-A1C	5.3		5.1	%	4.3～5.8
		47			
★白血球数	53	400	46	×10³/μl	36～92
★赤血球数	426	11.7	424	×10⁴/μl	380～500
★ヘモグロビン量	12.5	38.6	12.6	g/dl	11.2～14.
★ヘマトクリット値	40.7	97	40.8	%	33.5～43.
MCV	96	29.3	96	fl	81～98
MCH	29.3	↓ 30.3	29.7	pg	27.0～33.
MCHC	↓ 30.7	15.8	↓ 30.9	%	32.0～35.
★血小板数	22.5		21.7	×10⁴/μl	14.0～35.
		0.0			
		0.0	0.0	%	0.0
		51.1	0.0	%	0.0
		1.5	51.6	%	36.0～69.
		1.1	2.6	%	1.0～5.(
		41.4	1.8	%	0.0～2.(
		0.0	40.7	%	27.0～53.
		4.9	0.0	%	0.0
			3.3	%	2.0～10.

総合報告書Ⅰ	受付 11/03/0	受付 11/03/ 受付 11/03/30	報告 11/03/3
	登録衛生検査	登録衛生検: 登録衛生検査所	実施料 329
責任者	(株)メディッ	(株)メディ (株)メディック	
大谷 敦	TEL(059)2	TEL(059) TEL(059)224-5455	

検査成績報告

CONFIDENTIAL

検査成績報告書

3389(2377) 津健康クリニック [redacted] 様

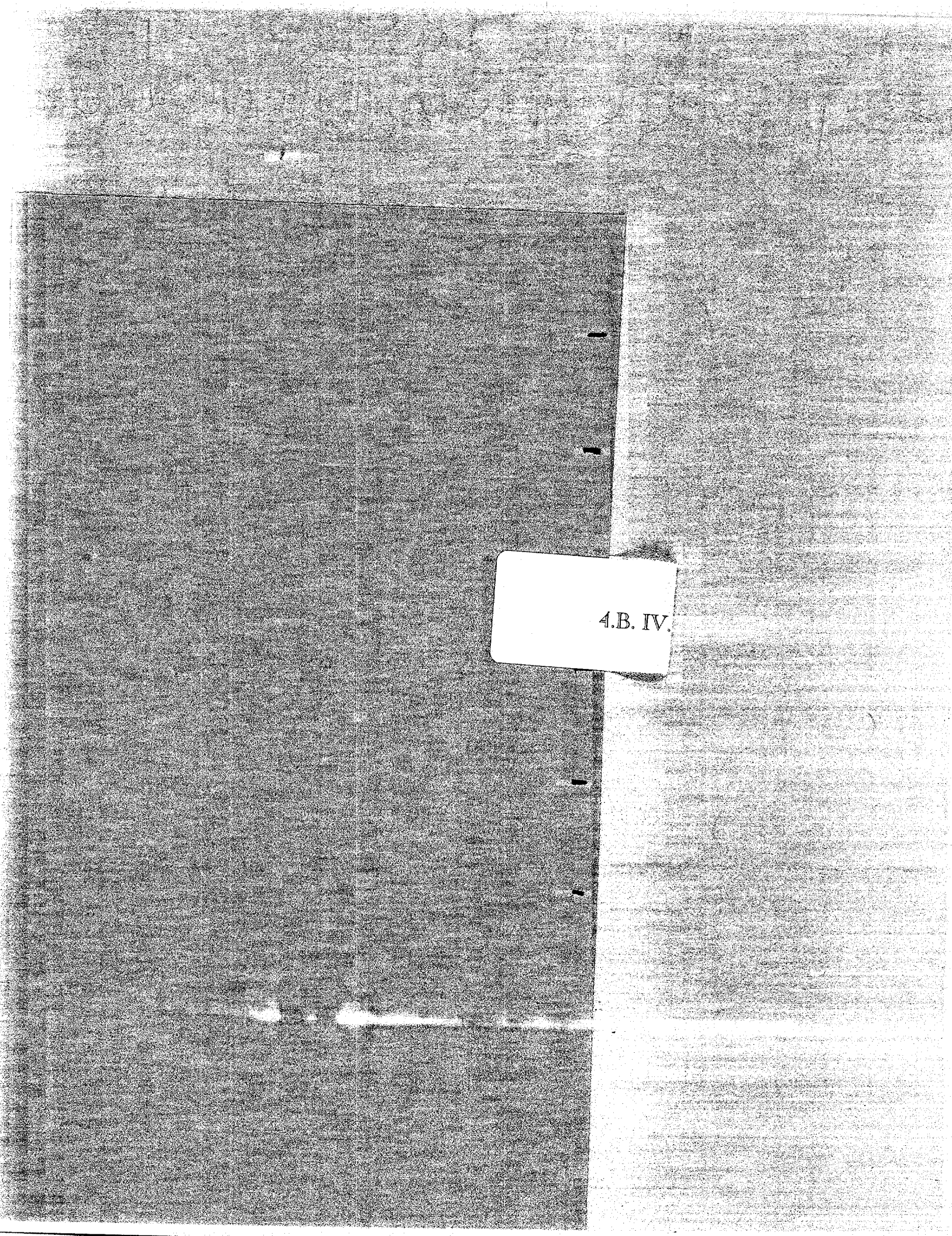
女性 65才

検査項目	測定値	測定値	測定値	検査項目	測定値	単位	基準値
		実施料 329点	実施料 244点		実施料 329点		
	コメント	ID№ 2-77003	ID№ 2-77084		ID№ 2-77090	コメント	
採取	受付 11/04/13	受付 11/04/27	受付 11/05/11	採取	受付 11/05/25	報告 11/05/26	
★血清総蛋白	7.6	7.7	7.8	*血清総蛋白	8.1	g/dl	6.5～8.2
★A/G比	1.4	1.1	1.3	*A/G比	1.5		1.1～2.0
★アルブミン	4.4	4.1	4.4	*アルブミン	4.8	g/dl	3.7～5.3
★ZTT	3.6	4.0	4.1	*ZTT	3.6	units	3.0～12.0
★ALP	180	181	203	*ALP	200	IU/l	100～350
★LAP	49	47	50	*LAP	48	IU/l	35～75
★GOT	20	19	18	*GOT	20	IU/l	10～40
★GPT	11	9	9	*GPT	13	IU/l	6～40
★LDH	259	263	295	*LDH	292	IU/l	230～460
★γ-GTP	9	7	10	*γ-GTP	11	IU/l	50以下
★総コレステロール	↗ 228	↗ 238	↑ 251	*総コレステロール	↗ 239	mg/dl	130～220
★トリグリセライド	↗ 157	↗ 223	↗ 180	*トリグリセライド	↗ 220	mg/dl	35～150
★尿素窒素	15	18	17	*尿素窒素	14	mg/dl	8～21
★クレアチニン	0.9	0.8	0.9	*クレアチニン	0.8	mg/dl	0.6～1.3
★尿酸	4.1	3.8	4.7	*尿酸	3.8	mg/dl	2.0～6.5
		5.5		*Hb-A1C	5.1	%	4.3～5.8
★白血球数	54		44				
★赤血球数	415	43	425	*白血球数	42	×10³/μl	36～92
★ヘモグロビン量	12.4	399	12.6	*赤血球数	419	×10⁴/μl	380～500
★ヘマトクリット値	39.1	11.9	40.5	*ヘモグロビン量	12.5	g/dl	11.2～14.7
MCV	94	37.6	95	*ヘマトクリット値	39.4	%	33.5～43.5
MCH	29.9	94	29.6	MCV	94	fl	81～98
MCHC	↘ 31.7	29.8	↘ 31.1	MCH	29.8	pg	27.0～33.5
★血小板数	19.2	↘ 31.6	21.6	MCHC	↘ 31.7	%	32.0～35.0
★血液像		19.8		*血小板数	21.3	×10⁴/μl	14.0～35.0
骨髄球	0.0		0.0	*血液像			
後骨髄球	0.0	0.0	0.0	骨髄球	0.0	%	0.0
好中球	54.4	0.0	56.2	後骨髄球	0.0	%	0.0
好酸球	1.5	62.4	1.8	好中球	53.8	%	36.0～69.0
好塩基球	0.9	1.9	1.1	好酸球	1.9	%	1.0～5.0
リンパ球	39.5	1.2	37.5	好塩基球	1.2	%	0.0～2.0
異型リンパ球	0.0	31.7	0.0	リンパ球	37.9	%	27.0～53.0
単球	3.7	0.0	3.4	異型リンパ球	0.0	%	0.0
大小不同		2.8		単球	5.2	%	2.0～10.0
環状				大小不同			
奇形				環状			
赤芽球				奇形			
				赤芽球			

総合報告書Ⅰ

受付	11/04/1	受付 11/04/27	受付 11/05/11	総合報告書Ⅰ	受付 11/05/25	報告 11/05/26
	登録衛生検査	登録衛生検査	登録衛生検査		登録衛生検査所	
責任者	(株)メディッ	(株)メディ	(株)メディ	責任者	(株)メディック	
大谷 敦	TEL(059)2	TEL(059)2	TEL(059)22	大谷 敦	TEL(059)224-5455	

[redacted]


4.B. IV.

Safety Test for Long-term Administration of Himematsutake [Iwade Strain 101]® Powder in Healthy Volunteers

Hitoshi. Ito, M.D., Ph. D.
Department of Pharmacology
Mie University School of Medicine
Edobashi, Tsu, Mie 514-0001
Japan
Research Institute of Mycology and Pharmacology
1-9 Suehiro-cho, Tsu, Mie 514-0012
Japan

Shiro Suzuki, M.D., Ph.D.
Tsu Health Clinic
799-7, Kannonji-cho, Tsu, Mie, 514-0062
Japan

Introduction:

Himematsutake (official name in Japanese), *Agaricus blazei* Murrill, is an edible mushroom introduced to the late Dr. Inosuke Iwade, professor of Forest Chemistry and Applied Mushroom Science of the Faculty of Agriculture, Mie University, Japan in 1965. Prof. Iwade founded a company named Iwade Research Institute of Mycology to analyze mushrooms chemically and conduct study about ingredients of mushrooms. In 1975, Iwade Research Institute of Mycology, with tremendous efforts by its staffs, succeeded in artificial cultivation of Himematsutake for the first time in the world.

With extensive study on Himematsutake led by Dr. Hitoshi Ito at Department of Pharmacology, Mie University School of Medicine, Japan, the research team discovered Himematsutake's strong antitumor effects. Then they identified one strain, which marked the highest antitumor effects, and named it "Himematsutake [Iwade Strain 101]®".

Himematsutake [Iwade Strain 101]® and its antitumor effects were tested on numbers of experiments and reported to the academic and scientific associations and meetings. In 1980, the research on antitumor effects of Himematsutake [Iwade Strain 101]® was officially introduced to the 39th Annual Meeting of the Japanese Cancer Association and the 54th General Meeting of Japanese Pharmacological Society. Since then, the research has been continuously conducted and reported to the reputable meetings. The discovery of antitumor effects of Himematsutake [Iwade Strain 101]® and its effectiveness on tumor-implanted mice captured tremendous people's attention and was exposed on mass mediums such as TV, newspapers and magazines in Japan.

Iwade Research Institute of Mycology established mass production of the mushroom in 1983. Over 10,000 people who tend to be cancer patients have administered the Himematsutake [Iwade Strain 101]® products in a form of dried, powder and granule. They usually take the amount of the product that is equivalent to 5g - 50g of fruiting body of dried mushroom per day. There has been no report of side effect for long-term administration of the products. Therefore, it is conceived that the Himematsutake [Iwade Strain 101]® products are the extremely safe edible mushroom.

In this report, the healthy volunteers, who agreed to cooperate with the theme of this study, were tested, with an open & trial method, to determine if the Himematsutake [Iwade Strain 101]® powder would be safe for long-term administration and/or there would be any side effect. The following is the report of the trial:

I. Material Sample:

The material sample, Himematsutake [Iwade Strain 101]® Powder was provided by Iwade Research Institute of Mycology at 1-9 Suehiro-cho, Tsu, Mie 514-0012, Japan.

Chemical Specifications of Himematsutake [Iwade Strain 101]® Powder:

Energy	Kcal/100g	350
Water Content	g/100g	1.2
Crude Ash	g/100g	1.2
Crude Protein	g/100g	7.0
Crude Fat	g/100g	0.6
Crude Fiber	g/100g	0.9
Total Sugar	g/100g	19.1
GS Fiber	g/100g	70.0

Remarks: 1 pack contains 5 g of Himematsutake [Iwade Strain 101]® Powder

II. Subject:

The subjects were 20 male and 15 female university students ranging in age from 19 to 23.

III. Methods and Terms of Administration:

For male, 30g (6 packs) of Himematsutake [Iwade Strain 101]® Powder per day, 2 packs after every meal - 3 times a day, were given for administration. For female, 15g (3 packs), 1 pack after every meal - 3 times a day, were given for administration.

The term of administration was for a period of 6 months.

Remarks:

- Originally, there were 25 male and 19 female subjects. However, 5 male and 3 female, who went back to their home town during the summer and winter vacation and stopped taking Himematsutake [Iwade Strain 101]® Powder, were excluded from the trial. Also, 1 female, who had other medication during the term was excluded as well.
- Double blind test was not proceeded for this trial.

IV. Clinical Test Items:

WBC, RBC, Hb, Hct, MCV, MCH, MCHC, Platelet, GOT, GPT, T-Chol., Triglyceride, HDL-C and Uric Acid were tested monthly on each subject for a period of 6 months. Each subject wrote down any signs and changes of her physical conditions on the questionnaire.

V. Result of Clinical Test:

The result was compared between male and female. As it is shown, the figures on RBC, Hb, Hct and Uric Acid for female were relatively lower than the ones for male. However, the figures were still all in a normal range.

There was no significant change on WBC, MCV, MCH, MCHC, Platelet, GOT, GPT, T-Chol., Triglyceride and HDL-C.

VI. Result of Subjective Signs and Improvement:

The result of subjective signs and improvement indicated on Table 3 & 4 were all claimed by the subjects.

Table 3 - Subjective Signs after taking Himematsutake [Iwade Strain 101] ® Powder for 20 male subjects

Subjective Signs		Before		1 month after		3 months after		6 months after	
		No.	%	No.	%	No.	%	No.	%
Appetite	(-)	3	15	1	5	0	0	0	0
	(±)	9	45	6	30	5	25	3	15
	(+)	8	40	13	65	15	75	17	85
Digestion	(-)	11	55	16	80	14	70	15	75
	(±)	6	30	4	20	6	30	5	25
	(+)	3	15	0	0	0	0	0	0
Stool (amount)	(-)	2	10	0	0	0	0	0	0
	(±)	13	65	5	25	6	30	6	30
	(+)	5	25	15	75	14	70	14	70
Urine (amount)	(-)	0	0	0	0	0	0	0	0
	(±)	13	65	8	40	7	35	6	30
	(+)	7	35	12	60	13	65	14	70
General Condition	(-)	2	10	0	0	0	0	0	0
	(±)	7	35	6	30	6	30	6	30
	(+)	11	55	14	70	14	70	14	70

Table 4 - Subjective Signs after taking Himematsutake [Iwade Strain 101] ® Powder for 15 female subjects

Subjective Signs		Before		1 month after		3 months after		6 months after	
		No.	%	No.	%	No.	%	No.	%
Appetite	(-)	5	33	3	20	1	7	1	7
	(±)	10	67	2	13	1	7	1	7
	(+)	0	0	10	67	13	87	13	87
Digestion	(-)	10	67	12	80	13	87	15	100
	(±)	3	20	2	13	2	13	0	0
	(+)	2	13	1	7	0	0	0	0
Stool (amount)	(-)	4	27	0	0	1	7	0	0
	(±)	8	53	4	27	2	13	3	20
	(+)	3	20	11	73	12	80	12	80
Urine (amount)	(-)	1	7	0	0	0	0	0	0
	(±)	12	80	6	40	5	33	4	27
	(+)	2	13	9	60	10	67	11	73
General Condition	(-)	3	20	0	0	0	0	0	0
	(±)	8	53	11	73	10	67	8	53
	(+)	4	27	4	27	5	33	7	47

VII. Side Effect:

There was no significant side effect observed or claimed. 2 male and 1 female subjects expressed that they did not prefer the taste and smell of the mushroom. However, the preference on the taste and smell of the mushroom varies depending on the person. Also, this point is referred to other edible mushrooms, and therefore, shall not be necessarily considered.

VIII. Conclusion:

The 20 male subjects consumed 30g and 15 female subjects consumed 15g of Himematsutake [Iwade Strain 101] ® Powder per day for a period of 6 months. As a result, no hematological and biochemical abnormality were found in the subjects. On the other hand, subjective improvements such as smoothie and larger amount of evacuation and urination, and better sight, increased after 2 weeks of administration. With these in mind, it is conceived that Himematsutake [Iwade Strain 101] ® Powder is safe for long-term administration and has no side effect.

Table.1 Changes of hematological and Biochemical findings in healthy volunteers treated orally with 30 g of Himematsutake Powder for 6 months

Case No. 1～20　　Age : between the ages of 19 and 23　　Sex : male

Case No. / Month	Normal range	n = 20	n = 20	n = 19	n = 20	n = 18	n = 20	n = 20
		0	1	2	3	4	5	6
WBC ($x10^2/\mu l$)	36～92	71.2±10.1	71.4±12.3	73.7±9.7	72.3±11.6	73.1±11.8	72.0±9.7	72.8±10.5
RBC ($x10^4/\mu l$)	420～560	470±20.2	489±25.6	494±30.7	521±26.9	511±33.1	498±24.6	511±25.8
Hb (g/dl)	13.0～17.0	16.2±0.51	16.4±1.57	15.7±0.46	16.3±1.63	16.1±1.10	15.8±0.56	16.0±0.89
Hct (%)	39.0～50.0	43±1.3	44±2.1	43±1.8	44±1.5	44±1.7	46±1.8	45±1.9
MCV (fl)	81～98	86.1±1.17	83.4±2.34	85.7±2.98	89.1±3.31	90.4±3.26	85.9±3.91	86.2±3.11
MCH (pg)	27.0～33.5	27.4±0.69	28.4±0.93	27.9±1.50	29.8±1.45	29.9±1.30	30.2±1.89	28.2±1.52
MCHC (%)	32.0～35.0	34.3±0.65	35.0±0.59	34.7±0.89	34.9±0.64	32.8±0.75	33.6±0.71	34.1±0.78
Plt ($x10^4/\mu l$)	14.0～35.0	24.6±1.21	25.5±1.00	25.3±1.73	26.7±2.53	27.9±2.70	28.4±3.43	27.0±3.78
GOT (IU/l)	10～40	30±2.9	27±4.1	29±3.2	32±3.7	31±2.8	28±2.6	30±3.6
GPT (IU/l)	3～30	16±1.8	13±2.0	14±2.6	20±6.9	21±5.5	19±2.3	17±2.9
T-CHO (mg/dl)	120～240	139±14.3	142±11.8	131±12.9	132±17.6	147±12.7	141±14.5	136±15.2
TG (mg/dl)	40～170	97±18.1	93±23.9	101±37.1	119±31.5	116±28.2	120±25.1	124±39.3
HDL-C (mg/dl)	30～75	51±8.2	48±10.1	47±9.3	48±6.9	56±5.8	42±9.5	50±12.8
Uric acid (mg/dl)	2.5～7.5	4.4±0.57	4.2±0.40	4.1±0.49	4.5±1.28	4.0±0.63	4.9±1.10	4.7±1.33

Values are expressed as means ± S.E.

Table. 2 Changes of hematological and Biochemical findings in healthy volunteers treated orally with 15 g of Himematsutake Powder for 6 months

Case No. 21～35　　Age : between the ages of 19 and 23　　Sex : female

Case No. / Month	Normal range	n = 15	n = 15	n = 15	n = 15	n = 13	n = 13	n = 15
		0	1	2	3	4	5	6
WBC ($x10^2/\mu l$)	36～92	69.3±10.9	71.0±11.5	72.3±11.4	71.5±9.7	72.0±12.5	71.3±9.8	73.4±8.7
RBC ($x10^4/\mu l$)	420～560	452±22.3	479±28.2	486±29.1	473±24.7	463±19.8	468±23.3	479±24.4
Hb (g/dl)	13.0～17.0	14.6±0.32	14.4±0.54	15.0±1.17	15.7±0.93	15.4±0.67	15.5±0.57	15.4±0.79
Hct (%)	39.0～50.0	40±1.4	42±1.9	41±1.8	43±2.0	42±2.1	41±1.3	40±1.5
MCV (fl)	81～98	81.4±2.23	84.9±1.71	84.2±2.11	83.6±1.91	83.5±1.85	85.5±2.24	83.1±1.76
MCH (pg)	27.0～33.5	28.5±0.72	26.9±0.77	26.9±0.81	29.0±0.94	29.2±0.98	28.7±0.85	27.4±0.74
MCHC (%)	32.0～35.0	33.4±0.84	34.1±0.92	34.6±0.99	33.8±0.78	35.0±1.21	34.7±0.95	33.6±1.00
Plt ($x10^4/\mu l$)	14.0～35.0	23.6±1.53	23.1±0.84	25.9±1.72	23.8±1.37	25.8±2.64	26.0±1.94	25.4±1.30
GOT (IU/l)	10～40	29±2.3	24±5.1	22±8.7	26±5.7	22±3.5	24±4.1	23±3.6
GPT (IU/l)	3～30	22±2.7	22±2.9	18±1.9	20±2.3	19±2.8	22±1.8	19±2.1
T-CHO (mg/dl)	120～240	132±18.2	139±15.3	133±17.9	141±27.7	149±20.3	138±29.1	135±26.4
TG (mg/dl)	40～170	107±29.1	103±23.6	111±46.2	128±60.3	125±40.2	112±31.7	124±28.6
HDL-C (mg/dl)	30～75	50±9.6	53±6.4	52±7.6	58±8.2	54±6.5	56±7.2	62±9.8
Uric acid (mg/dl)	2.5～7.5	3.2±0.95	3.5±0.91	4.0±0.54	3.6±0.93	3.4±0.66	3.3±1.01	3.4±0.74

Values are expressed as means ± S.E.

4.B. V.

Clinical Trial with Himematsutake [Iwade Strain 101]® Powder

on Patients with Malignant Tumor

(Study on Long-term Administration and Side Effect)

Shiro Suzuki, M.D., Ph.D.
Tsu Health Clinic
799-7, Kannonji-cho, Tsu, Mie, 514-0062
Japan

Hitoshi. Ito, M.D., Ph. D.
Department of Pharmacology
Mie University School of Medicine
Edobashi, Tsu, Mie 514-0001
Japan
Research Institute of Mycology and Pharmacology
1-9 Suehiro-cho, Tsu, Mie 514-0012
Japan

Introduction:

In recent years, chemotherapy and immunotherapy have been taking a big role on cancer treatment, and numbers of clinical cases for those methods greately increased around the globe. On the other hand, Himematsutake (official name in Japan), one of edible mushrooms - scientific name: *Agaricus blazei* Murrill, has captured great attention from an immunotherapy field of view.

During this trial, we had an opportunity to review Himematsutake [Iwade Strain 101]® Powder on patients with malignant tumor to determine if the product would be safe for long-term administration and/or there would be any side effect. The following is the report of the clinical trial:

I. Material:

"Himematsutake [Iwade Strain 101]® Powder" was provided by Iwade Research Institute of Mycology Co., Ltd. at 1-9 Suehiro-cho, Tsu, Mie 514-0012, Japan.

Chemical Specifications of Himematsutake [Iwade Strain 101]® Powder:

Energy	kcal/100g	350
Water Content	g/100g	1.2
Crude Ash	g/100g	1.2
Crude Protein	g/100g	7.0
Crude Fat	g/100g	0.6
Crude Fiber	g/100g	0.9
Total Sugar	g/100g	19.1
GS Fiber	g/100g	70.0

Remarks: 1 pack contains 5 g of Himematsutake [Iwade Strain 101]® Powder

II. Subject Patients: (see Table 1)

Name of diagnosed cancer:

1) 1 case of uterine sarcoma
2) 2 case of ovarial tumor
3) 7 cases of cervical carcinoma Total 10 cases

Type of cancer:

1) 2 cases of cystadenoma
2) 7 cases of squamous cell carcinoma
3) 1 case of sarcoma

III. Treatment Methods

Each patient (except cases No.9 and 10) previously had a surgical operation before taking Himematsutake [Iwade Strain 101]® Powder. 2 packs-10g of Himematsutake [Iwade Strain 101]® Powder was given to each patient 3 times a day before every meal.

The total intake amount of Himematsutake [Iwade Strain 101]® Powder per person was between 3,360g (case No.1: 112days) and 10,860g (case No.5: 362days).

IV. Terms:

The term of administration was between 112 days and 362 days.

V. Other Conditions:

1 case had 5-FU treatment, 10mg/kg once a week for 10 consecutive weeks, before taking Himematsutake [Iwade Strain 101]® Powder.

In case of radiation therapy, Linac 6MV x-ray, ^{60}Co- γ -ray was given for 3 weeks, 200 rad per day, 1000 rad per week.

VI. Clinical Test Items:

1) Blood Test: RBC, Hemoglobin, Hematocrit, WBC, Platelet, Total Protein, Albamin/Globumin Ratio, γ -globulin, GOT, GPT

2) Delayed Skin Test (PPD): An intradermal injection of PPD - 0.05 μ g/0.1ml was given. The test result was conducted 48 hours after the injection. The average of vertical and horizontal lengths of erythema was measured.

   PPD - manufactured by Japan BCG

3) Intradermal PHA Test: An intradermal injection of the purified PHA - 5 μ g/0.1ml was given. The test result was conducted 24 hours after the injection. The average value of vertical and horizontal lengths of erythema was measured.

   Purified PHA - manufactured by Wellcome

4) Lymphocytes: Lymphocytes count in peripheral blood was measured by Hamatrak 360 Automated Differential System.

   Hamatrak 360 Automated Differential System - manufactured by Geometrac

   * As for NK cell, used Flow Cytometry method for measurement

VII. Results: (see Fig. 1 ~ 14)

1) RBC: (see Fig. 1)
There was no significant change. Instead, there was a tendency of increase.

2) Hemoglobin: (see Fig. 2)
There was no significant change.

3) Hematocrit: (see Fig.3)
No depression of hematocrit was observed.

4) WBC: (see Fig. 4)
The changes of WBC were observed, but there was no decrease.

5) Lymphocytes: (see Fig. 5)
Lymphocytes count was shown on the diagram as an index of immunotherapy. As the term of intake goes longer, lymphocytes count increased.

6) Platelet: (see Fig. 6)
There were slight changes, but no major changes were observed

7) Total Protein: (see Fig. 7)
There were slight changes, but no decrease was observed

8) Albumin/Globumin Ratio (see Fig. 8)
Observed a significant increase on case No. 9.

9) γ -globulin: (see Fig. 9)
Observed no significant change.

10) GOT: (see Fig. 10)
Observed remarkable depression on case No. 3, 9 and 10 whose GOT was high before taking Himematsutake [Iwade Strain 101]® Powder.

11) GPT: (see Fig. 11)
Observed depression on case No. 3, 9 and 10 whose GPT was relatively high before taking Himematsutake [Iwade Strain 101]® Powder.

- Observed no abnormal indication of GOT and GPT on all the subject patients after taking Himematsutake [Iwade Strain 101]® Powder.

12) PPD Response: (see Fig. 12)

Erythma (mm)		Before n=10	After 3M n=10	After 6M n=8
0 - 9	(-)	2	0	0
10 - 19	(+)	4	3	2
20>	(++)	4	7	6

Observed positive PPD Response on each subject patients. 2 subject patients marked negative PPD Response before taking Himematsutake [Iwade Strain 101] ® Powder, but turned to be positive (+ & ++) after taking it. As a result, all 6 subject patients notably increased their PPD Response even more toward positive after taking it.

13) PHA Response: (see Fig. 13)

Tested PHA Response on 6 subject patients - case No. 1, 2, 3, 7, 8 and 9 whose mean diameter of erythema was less than 20 mm on PPD Response.

2 out of 6 subject patients marked negative PHA Response when started taking Himematsutake [Iwade Strain 101]® Powder, but turned to be positive after taking it. As a result, all 6 subject patients, except No. 8, notably increased their PHA Response even more toward positive after taking it.

14) NK cell Response (see Fig. 14)

Tested NK cell Response on 6 subject patients - case No. 1, 2, 3, 7, 8, and 9. 2 out of 6 subject patients marked less than 10% of NK cell in peripheral blood while the normal range is 18% - 40%. After taking Himematsutake [Iwade Strain 101]® Powder, observed the remarkable increase of NK cell activity on all 6 subject patients.

VIII. Confirmed Benefits from Clinical Trial:

It is a common practice and procedure to evaluate this type of clinical trial without controlled subjects.

However, after 2 weeks of administration, all 10 subject patients experienced and confirmed the self-defined common physical changes such as improvements on general physical condition, taste better in food, hold tenseness on tummy, smooth stool and so on.

As far as the long-term administration is concerned, there was no side effect observed on 10 subject patients throughout the trial. As they took Himematsutake [Iwade Strain 101]® Powder for a longer period, clearly observed that they looked well and improved their skin condition.

IX. Conclusion:

1) As shown in the result of PPD and PHA Skin Test, it was confirmed that the immune system of all the subject patients (except case No. 8 in PHA skin test) was strengthened by the administration of Himematsutake [Iwade Strain 101]® Powder. This result shall support the result of the animal experiment, which was conducted previously. (see References 1-4)

2) As it is shown in the result, the trend of improvement on Lymphocytes count and NK cell activity were seen. This result shall support the result of the animal experiment, which was conducted previously.

* Based on the above 1) & 2), it is certain that Himematsutake [Iwade Strain 101]® Powder promotes cellular immunity response.

3) For those who had the high figures in GOT and GPT encountered the trend of decrease in them as indicated in the test result.

4) RBC was not significantly changed, but rather increased in a certain degree.

5) Even after the long-term administration, WBC remained within a normal range, and did not encounter any decrease during the trial.

6) Hemoglobin, hematocrit, platelet, total protein, A/G ratio and γ-globulin were all within a normal range during the trial

Based on the result from the trial, it is concluded that Himematsutake [Iwade Strain 101]® Powder is safe for long-term oral administration and causes no side effect. Furthermore, Himematsutake [Iwade Strain 101]® Powder can be the optimum adjuvant immunochemotherapy for a long-term use together with the series of treatment of surgical operation, chemotherapy and/or radiotherapy.

References:

1) Shimura K, Ito H and Hibasami H: Screening of host-mediated antitumor polysaccharides by crossed immunoelectrophoresis using fresh human serum, *Jpn J Pharmacol* 33, 403-408 (1983)

2) Itoh H, Ito H, Amano H and Noda H: Inhibitory action of a (1→6)-β-D-glucan-protein complex (F III-2-b) isolated from *Agaricus blazei* Murill ("Himematsutake") on Meth A fibrosarcoma-bearing mice and its antitumor mechanism. *Jpn J Pharmacol* 66, 265-271 (1994)

3) Ito H, Shimura K, Itoh H and Kawade H: Anti-tumor effects of a new polysaccharide-protein complex (ATOM) prepared from *Agaricus blazei* (Iwade Strain 101) "Himematsutake" and its mechnisms in tumor-bearing mice. *Anticancer Res* 17, 277-284 (1997)

4) Ito H: New Initiatives in Mycological Research Proceedings of the Third International Symposium of the Mycological Society of Japan. pp. 11-16 (1995)

Table. 1 Oral administration of Himematsutake [Iwade Strain 101] ® Powder on patients with malignant tumor

Cases No.	Age	Diagnosis	Treatment prior to Himematsutake [Iwade Strain 101] ®Powder			Treatment* (days)	Outcome after use
			chemotherapy	radition	surgical		
1	51	ut. Sarcoma	5-FU		ope	112	alive
2	48	OV. K1 (Cystadenoma)		Linac	ope	185	alive
3	53	OV. K (Cystadenoma)			ope	123	alive
4	49	C.C. I b (Squamous cell carcinoma)		Linac	ope	265	alive (rest)
5	59	C.C. II b (Squamous cell carcinoma)		Co	ope	362	alive (rest)
6	57	C.C. I b (Squamous cell carcinoma)		Co	ope	195	alive
7	40	C.C. II b (Squamous cell carcinoma)		Linac	ope	202	alive (recur)
8	62	C.C. II b (Squamous cell carcinoma)		Linac	ope	219	alive (rest)
9	41	C.C. I b (Squamous cell carcinoma)				227	alive
10	56	C.C. I b (Squamous cell carcinoma)		Linac		190	alive

* Himematutake [Iwade Strain 101]® Powder was given a certain period oral administration from 5-7 days after surgical operation

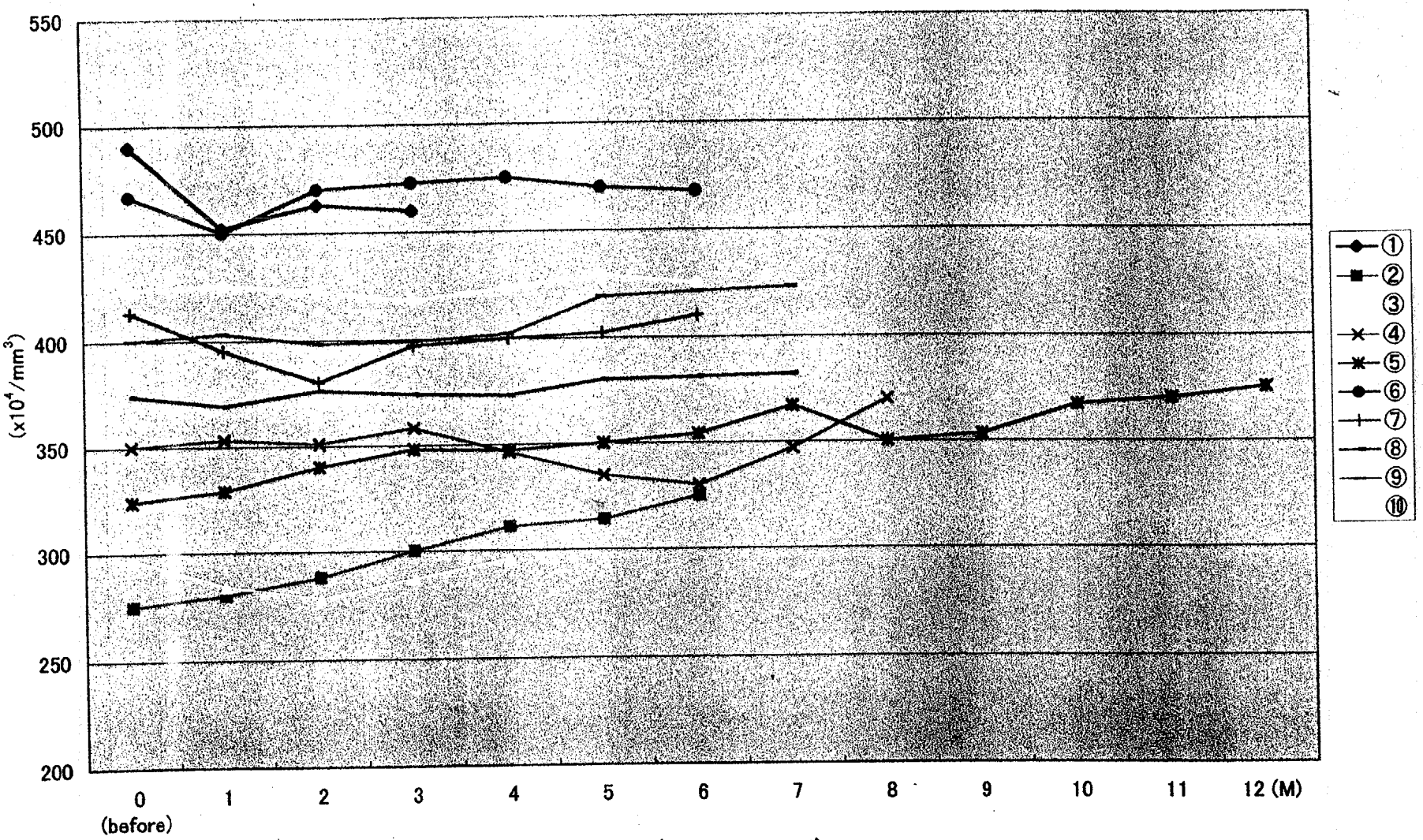


Fig. 1. RBC (red blood cell) count

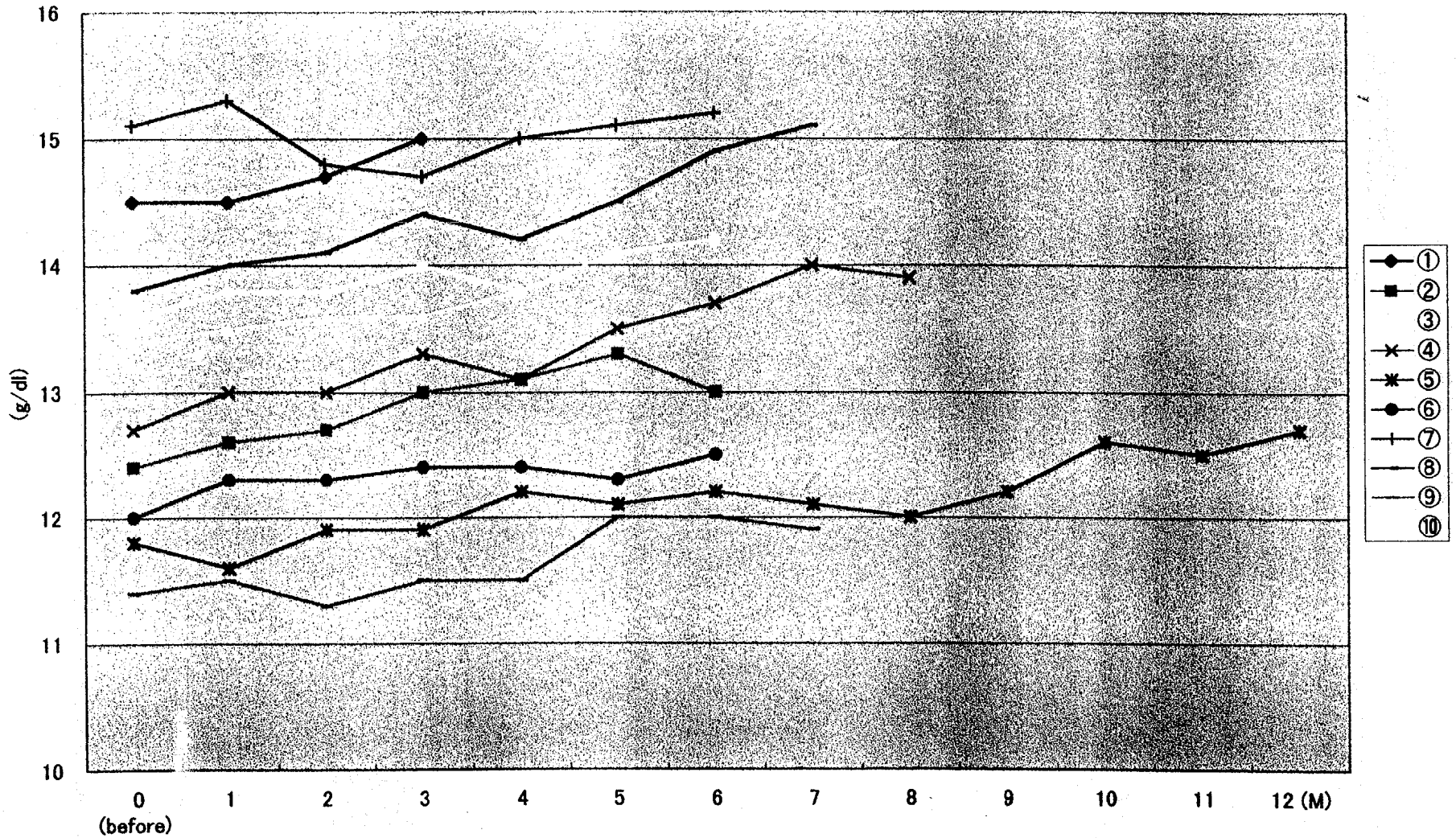


Fig. 2. Hb (hemoglobin)

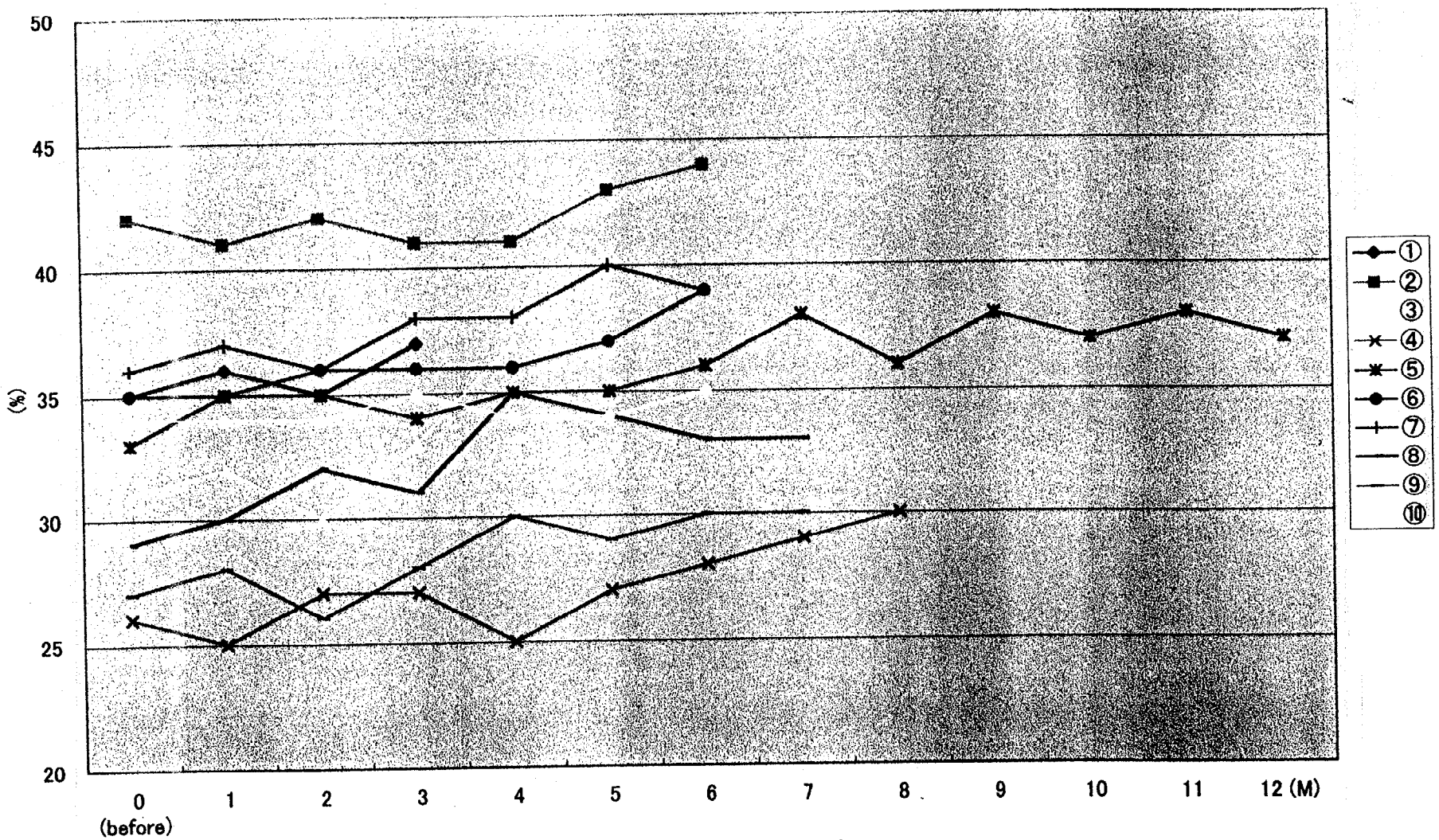


Fig. 3. Hct (hematocrit)

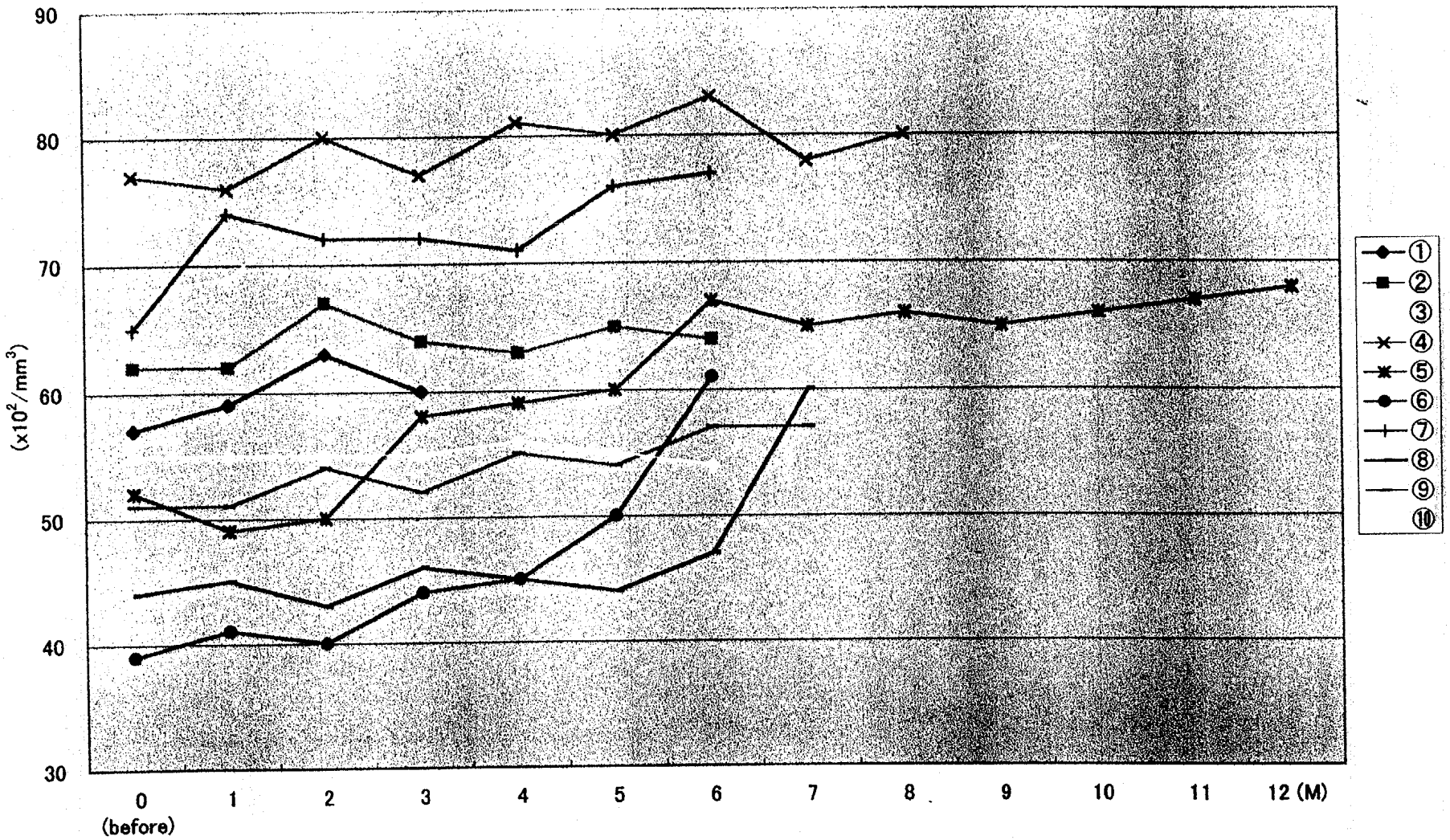


Fig. 4. WBC (white blood cell) count

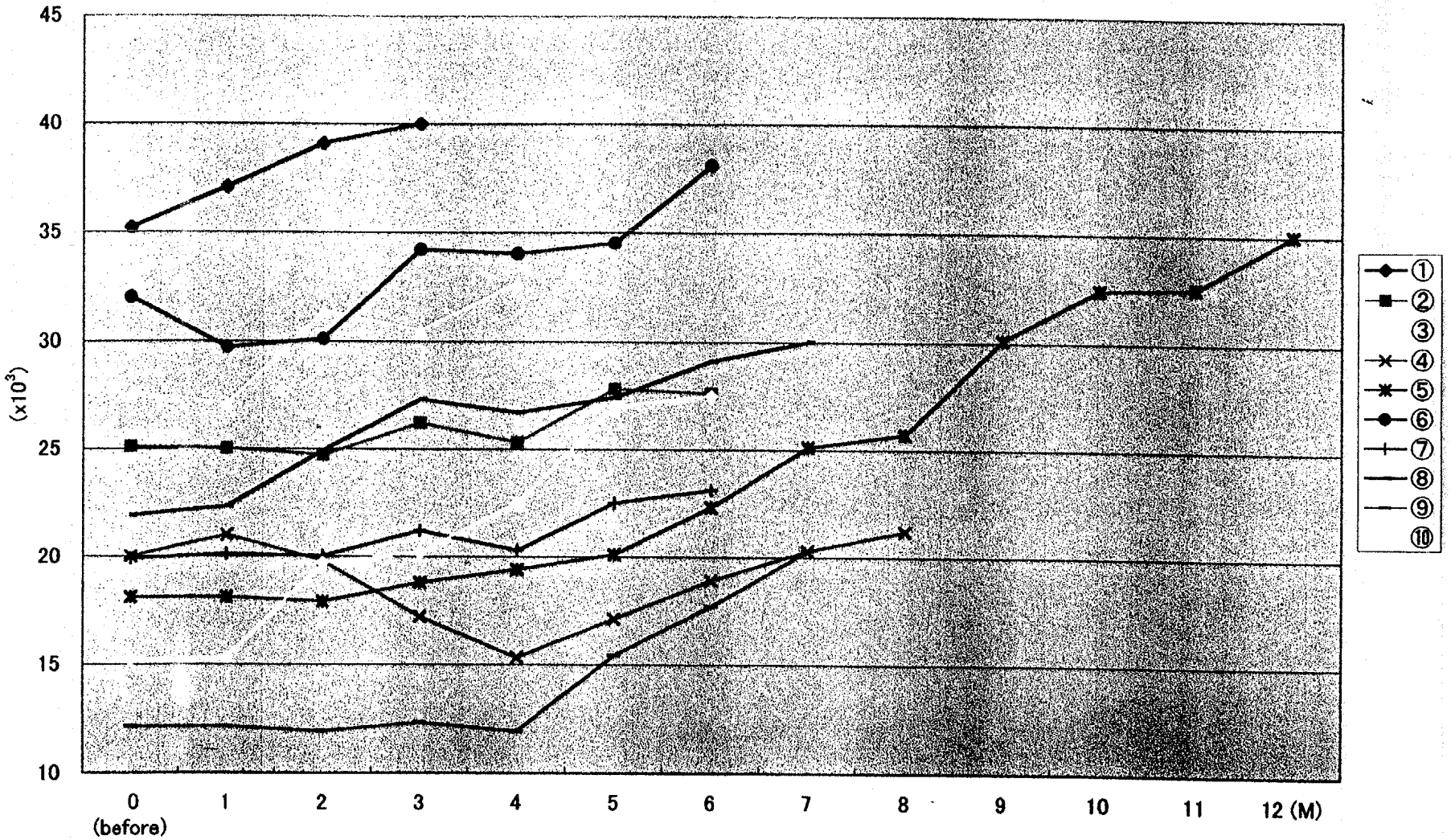


Fig. 5. Lymphocytes

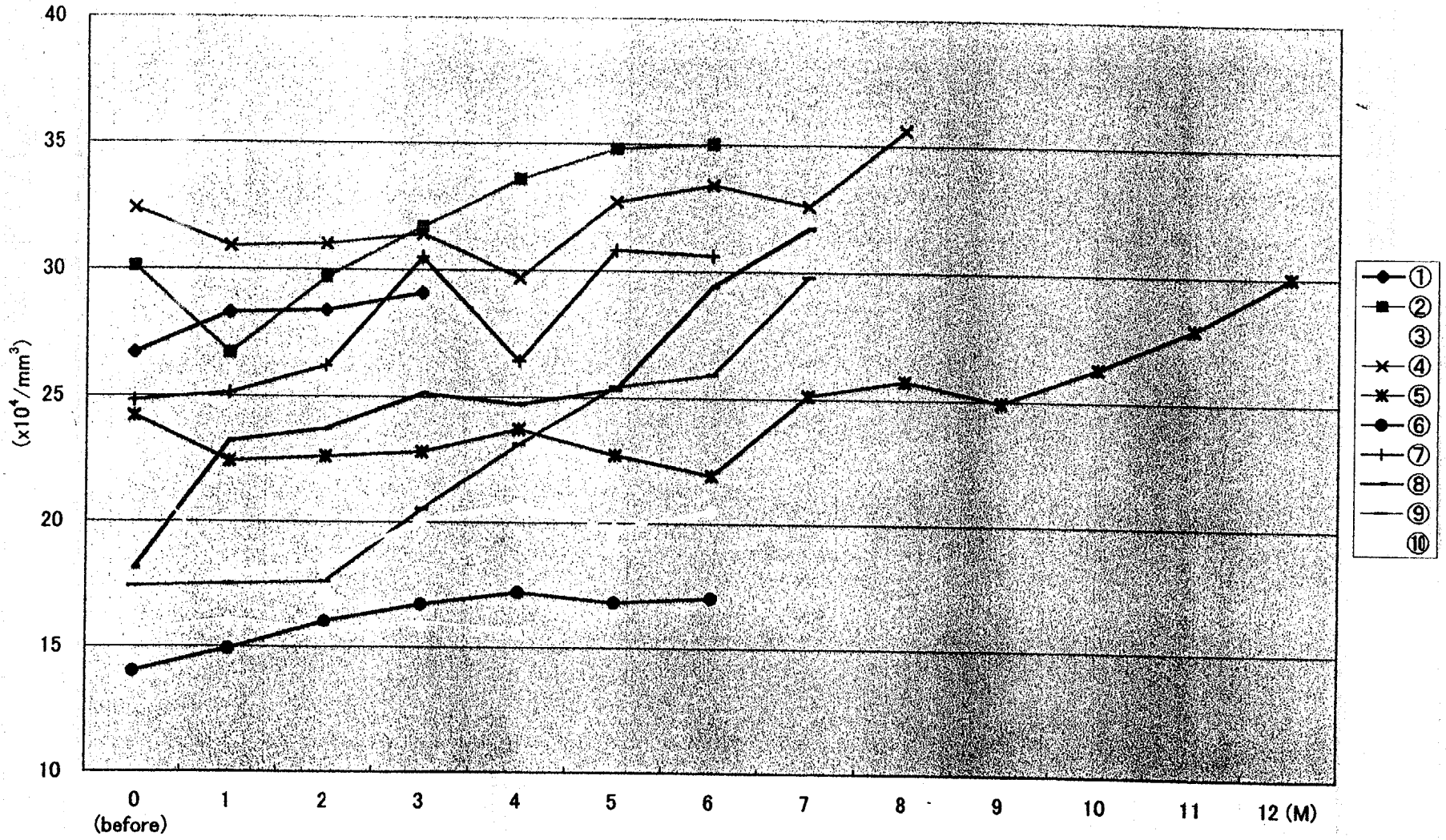


Fig. 6. Platelet

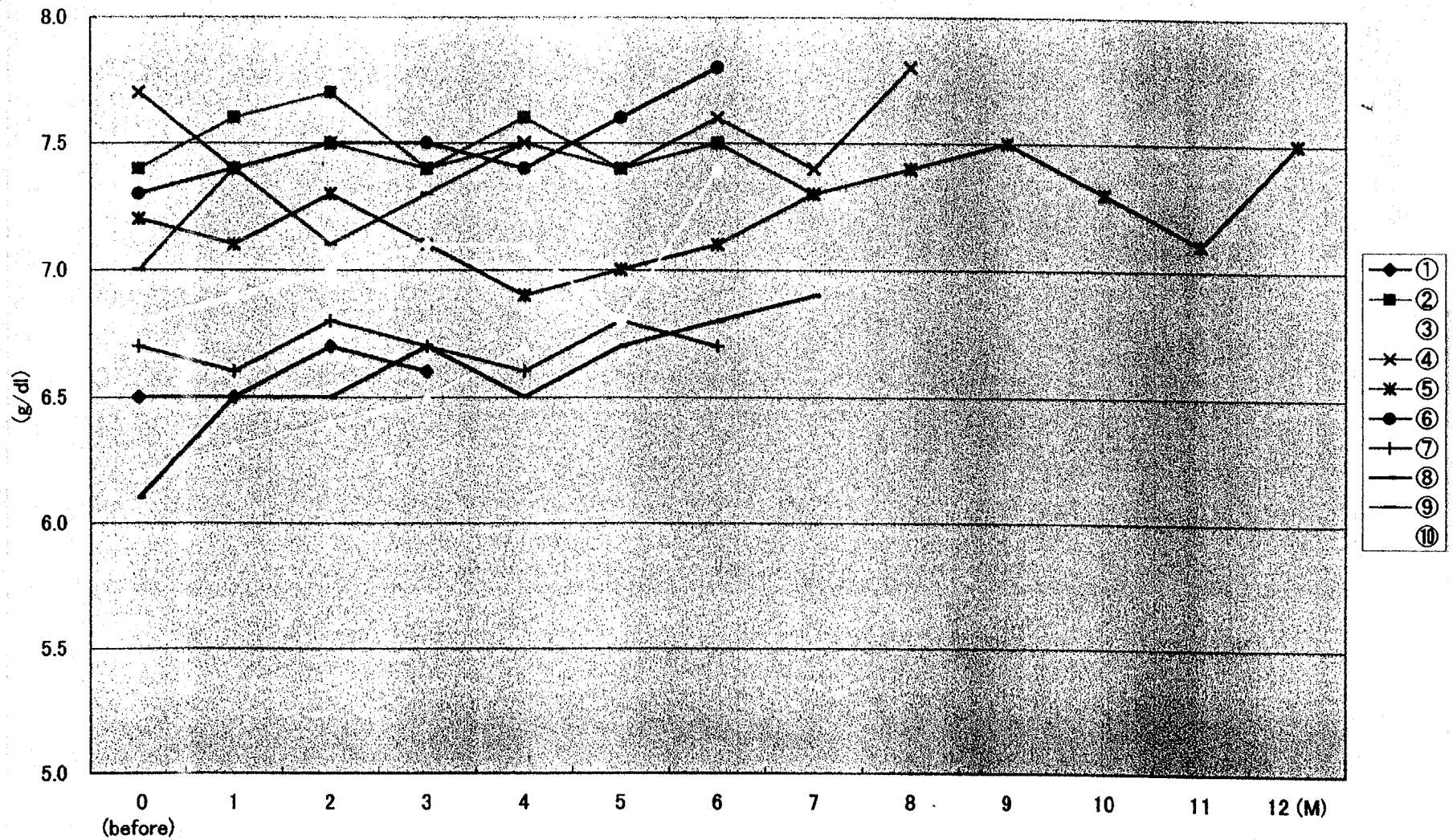


Fig. 7. Total Protein

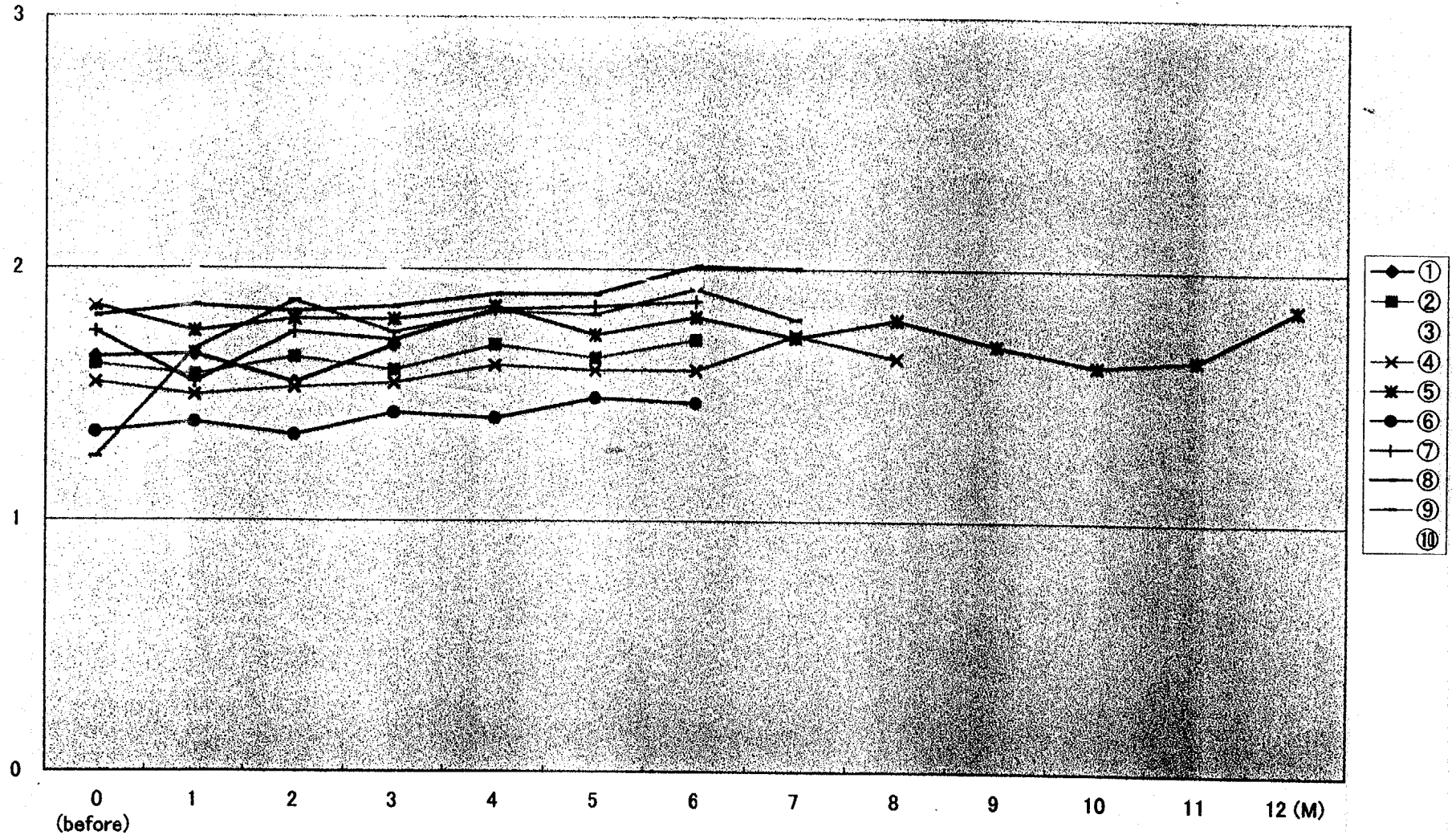


Fig. 8. A／G ratio

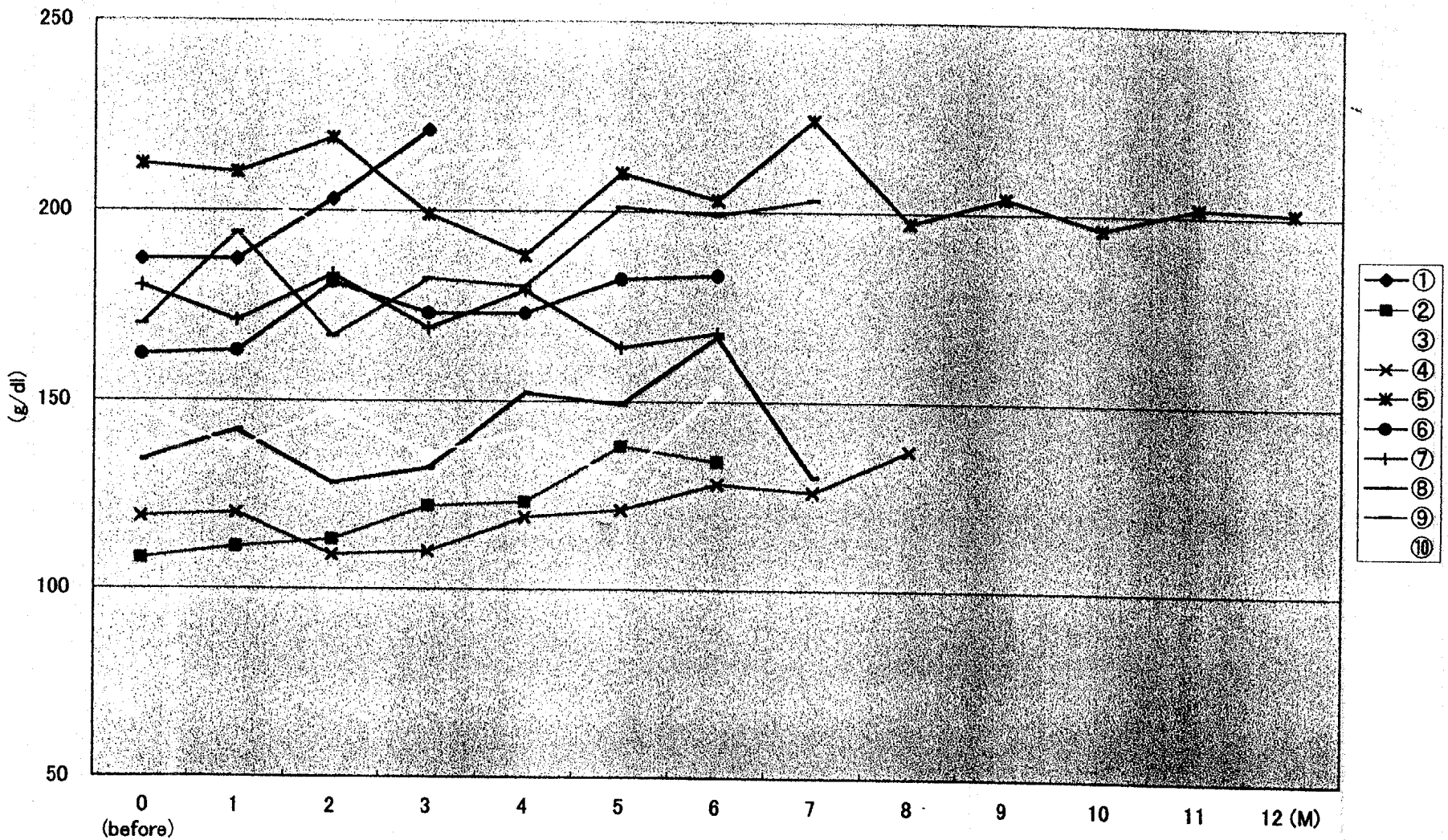


Fig. 9. γ—globurin

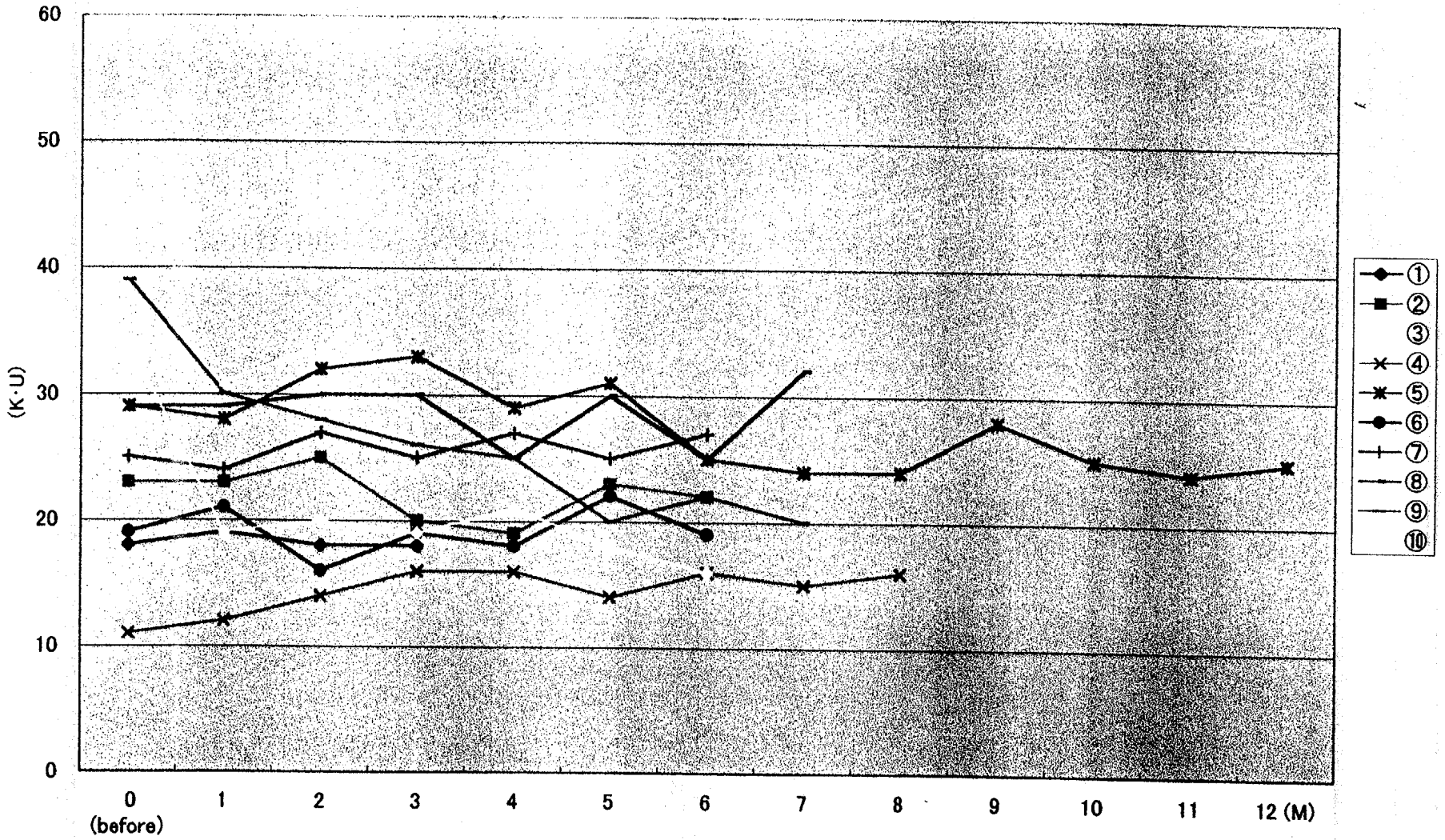


Fig. 10. GOT

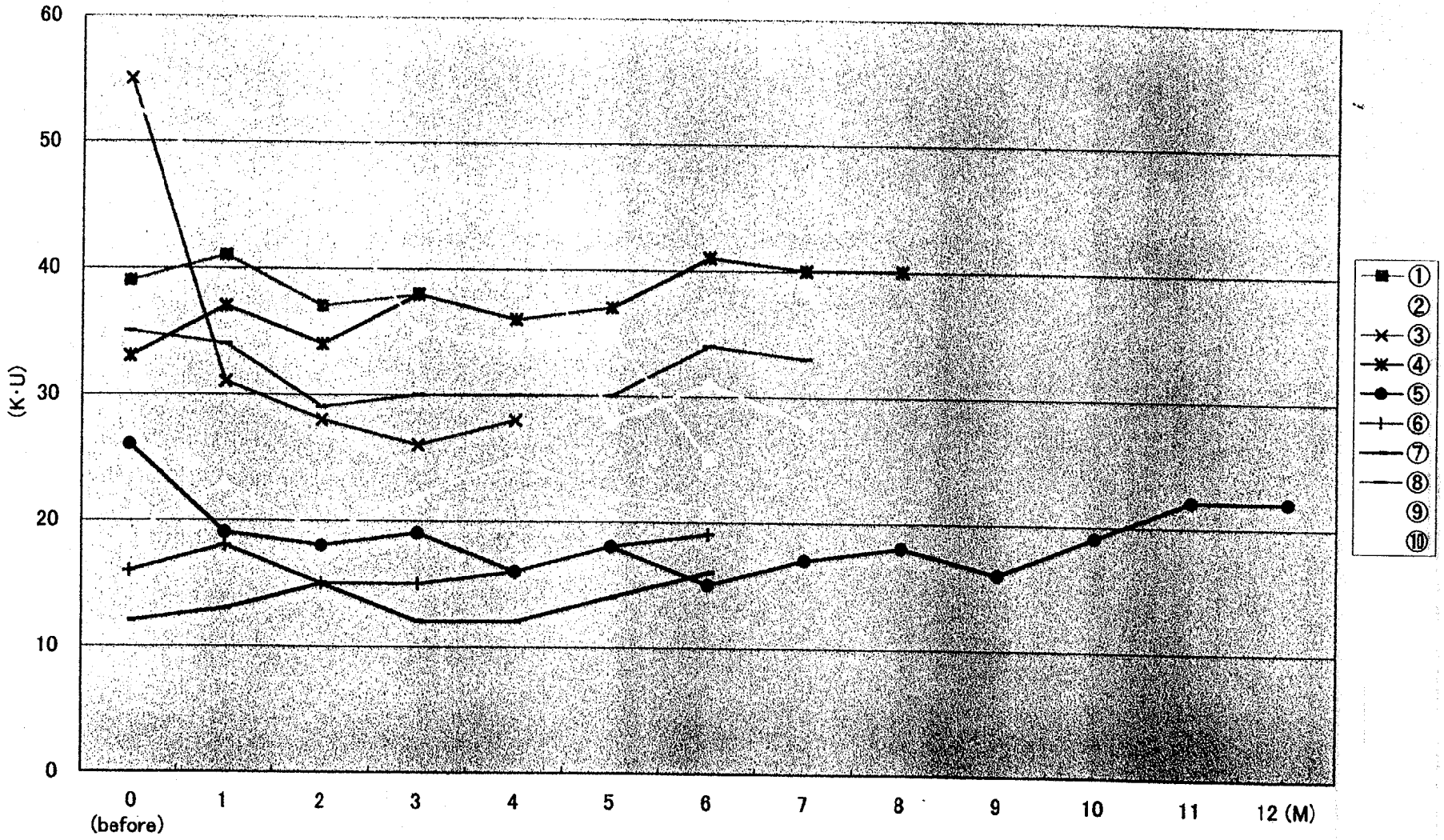


Fig. 11. GPT

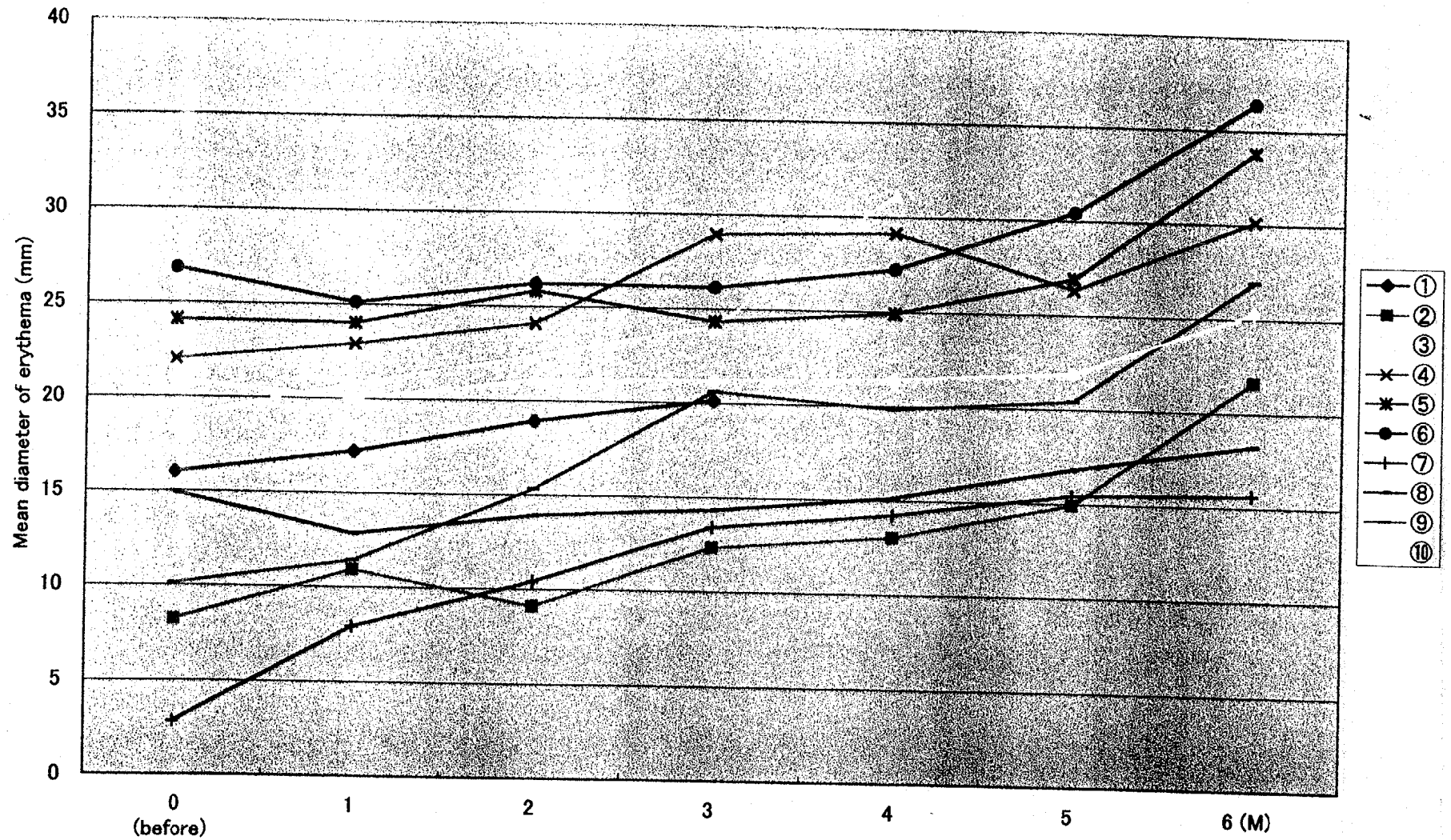


Fig. 12. Changes of PPD respones after treatment with Himematsutake Powder

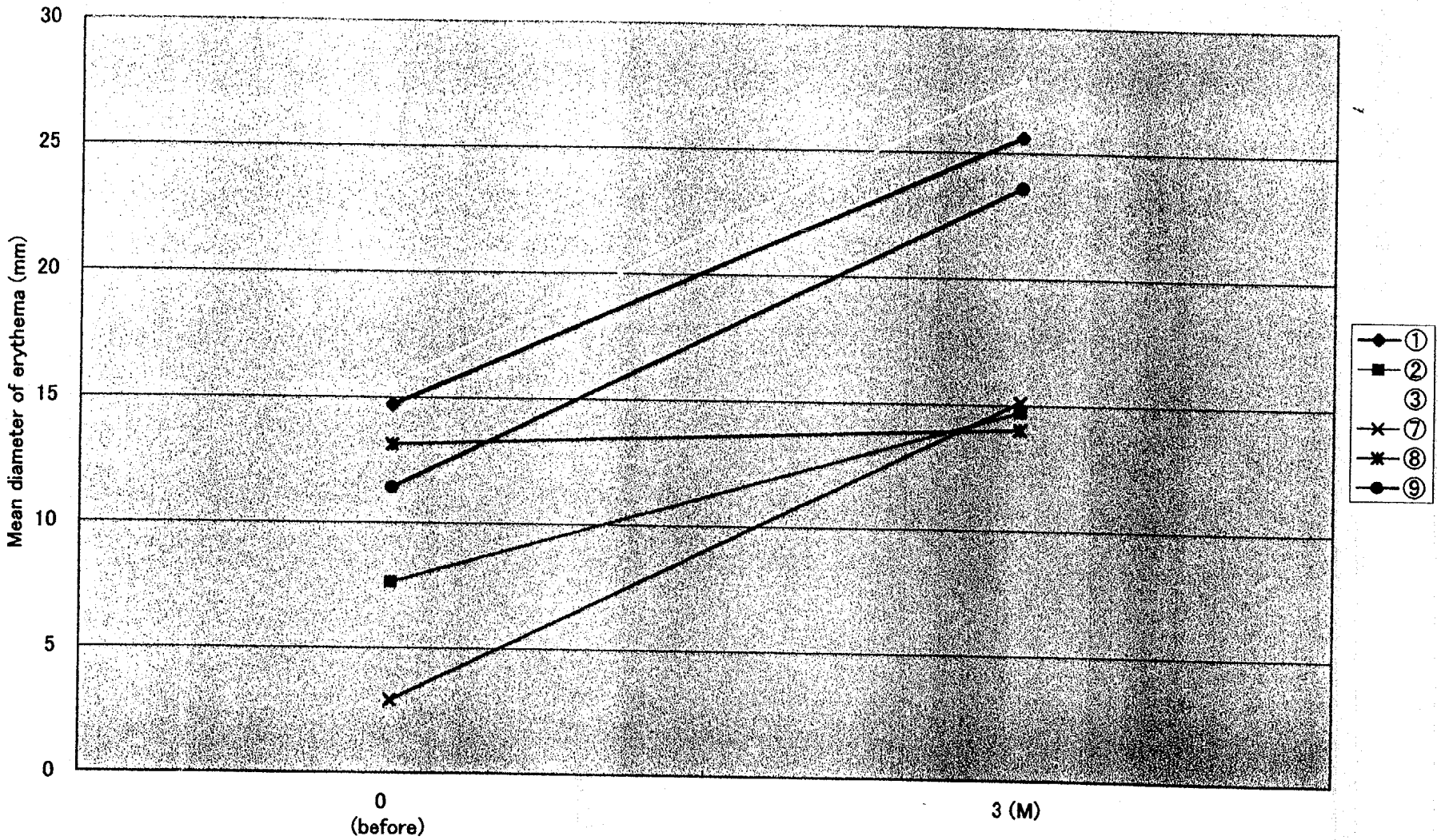


Fig. 13. Changes of PHA respones after treatment with Himematsutake Powder

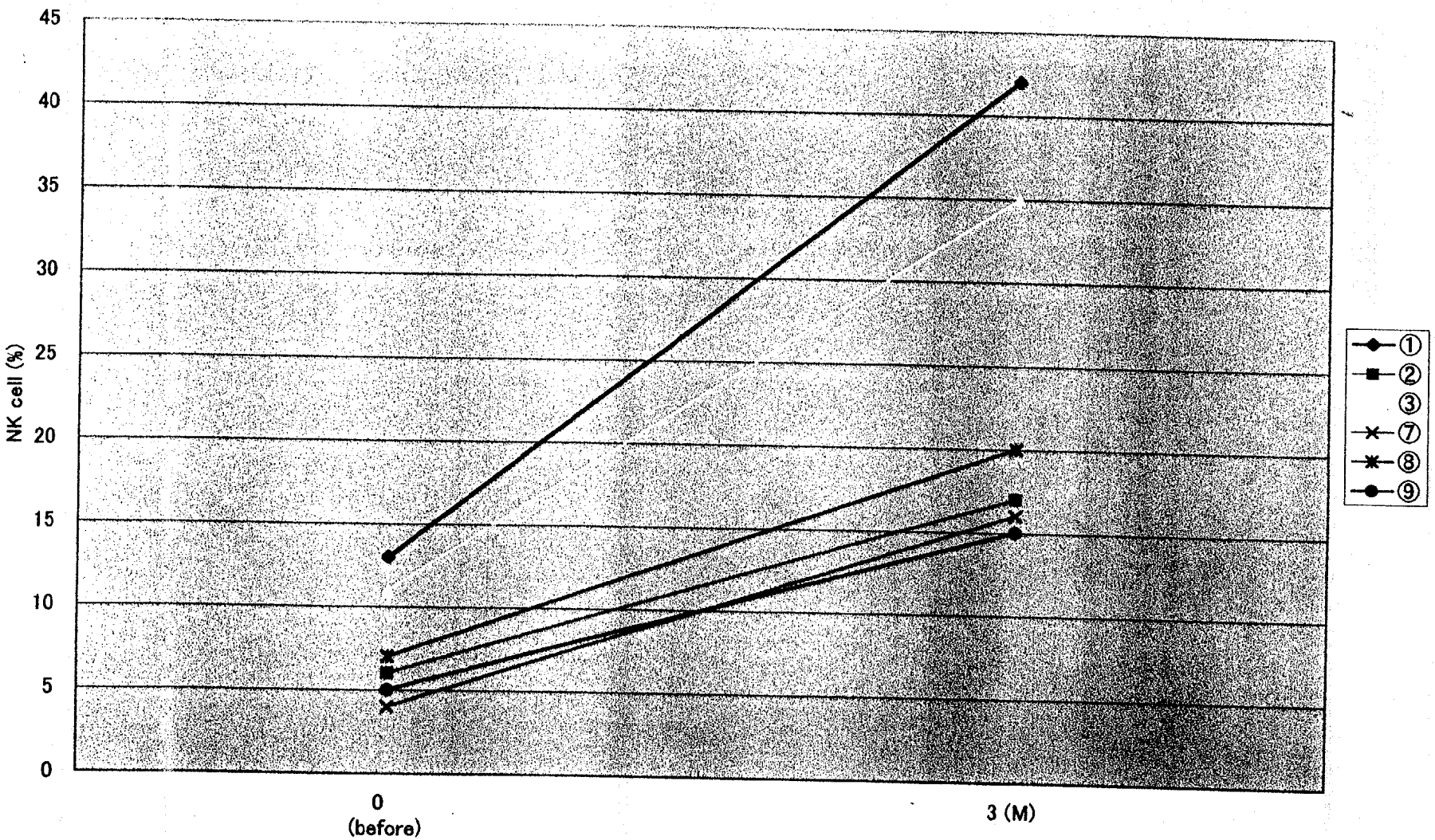
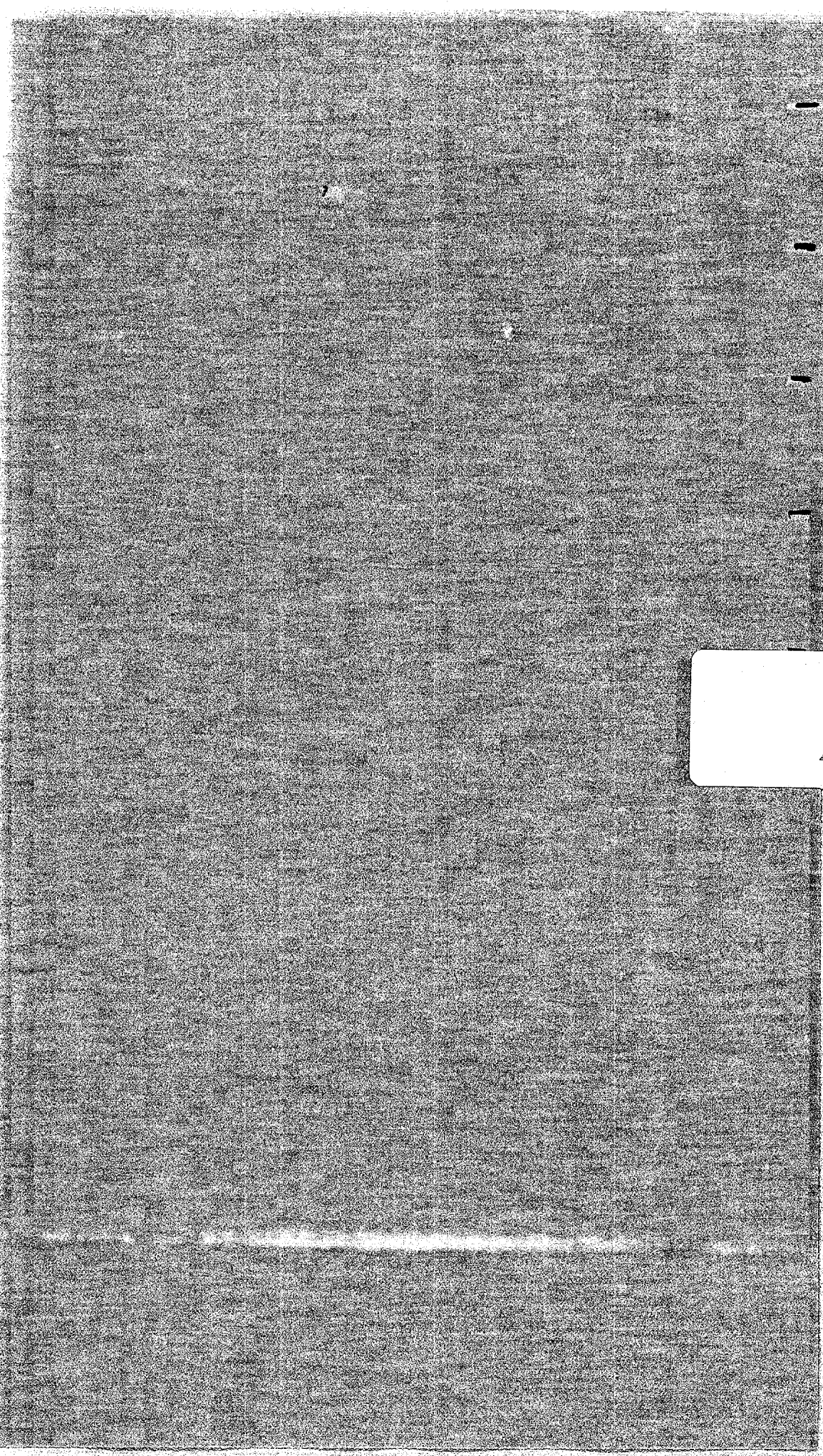


Fig. 14. Changes of NK cell respones after treatment with Himematsutake Powder



4.B.VI. **Revised Product specifications of Himematsutake Powder (CONFIDENTIAL)**